THE EFFECT OF N SOURCE, N RATE AND A VESICULAR-ARBUSCULAR MYCORRHIZAL FUNGUS ON NITROGEN LEACHING AND ELEMENTAL COMPOSITION IN BERMUDAGRASS TURF

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ABSTRACT

A glasshouse experiment was conducted to determine the effect of two sources and three rates of N and a mycorrhizal fungus on nitrogen leaching through common bermudagrass (Cynodon dactylon (L.) Pers.) grown on crushed basalt. Growth parameters were also measured to determine responses of common bermudagrass to N source, N rate and mycorrhizal infection.

More NO_3-N was leached from calcium nitrate than from ammonium sulphate. N lost as nitrate was 23%, 23% and 35% of N applied for the 24.4, 48.8 and 73.2 kg N/ha rates respectively of calcium nitrate. N lost as nitrate from ammonium sulphate was 21%, 12% and 10% of the N applied for the 24.4, 48.8 and 73.2 kg N/ha rates respectively.

There was no effect of mycorrhiza on leaching loss of nitrogen.

Root dry weights of mycorrhizal plants averaged 18854 kg/ha while non-mycorrhizal plants averaged 9819 kg /ha. Root dry weights were greater when ammonium sulphate was the source of N than with calcium nitrate.

With increasing levels of N, there were increasing concentrations of N, K and Mg in the clippings. Ca concentration of clippings decreased with increasing ammonium sulphate levels.

Silicon uptake was enhanced by mycorrhizal fungi. Higher concentrations were found in both roots and shoots of mycorrhizal plants.

VAM infection levels decreased with increasing levels of ammonium sulphate and remained the same with increasing levels of calcium nitrate. VAM infection levels also decreased with increasing depths. Higher average VAM infection levels with ammonium sulphate indicates a possible preference by the fungus for ammonium over nitrate.

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CHAPTER I

INTRODUCTION

Nitrogen (N) has long been known to be the most critical element in the maintenance of high quality turfgrass. It is required in the largest amount of any of the elements applied in fertilizer and therefore can be very costly. Its rate of movement through soil varies with the nitrogen source. Soluble N sources are used quite often in highly maintained turf because they are relatively inexpensive compared to other N sources and because once dissolved in the soil solution, they become readily available creating a rapid response in the plant. However soluble N sources, especially nitrate types, leach readily so that recovery of N by the plant may be low.

Rate of N movement through soil also depends on the physical characteristics of the medium itself. Sandy soils hold 8.3 cm or less of water per m of depth whereas finer-textured silty-clay loam can hold 21.0 cm per m (Aldrich, 1980). Thus, since certain forms of N move freely with the soil water and are not held by soil colloids, sandy soil will not retain N in the root zone well.

In the construction of golf putting greens, a stable sandy mix is desired to minimize compaction and runoff and to allow for high aeration porosity and consequently healthy root growth.

In the past, beach sand ($CaCO_3$) was the primary material used for building golf greens and amending soils in Hawaii because it was readily available in large supply. Beach sand however does not have good amending qualities because it eventually breaks down and releases large amounts of calcium which can create an alkaline condition and associated nutritional problems. This fact, along with the recent passage of state laws prohibiting the removal of beach sand from shoreline areas, has created a need for a new amending material.

In this study, crushed basaltic rock was used as a possible substitute for beach sand. Basalt is a major component of the volcanic islands of Hawaii and is therefore readily available. Basaltic rock is crushed into sand sized particles by a cement company. After crushing, it is screened into various size classes. It is currently being used most in the construction industry. A few golf courses have used it in building a limited number of greens. It is, therefore, necessary to know how rapidly various forms of N move and how much N is lost to leaching through this material so that an efficient fertilizer program can be established.

The influence of mycorrhizal fungi on nutrient uptake by host plants has been studied and is often considered important (Mosse, 1957; Gray and Gerdemann, 1973; Gerdemann, 1975). However little is known of the effect of mycorrhizae on the downward movement of nitrogen through soil. Haines and Best (1976) reported a reduction of NH_4 and NO_3-N loss through soil columns with Liquidambar styraciflua L. seedlings inoculated with Glomus mosseae (Nicol. and Gerd.) Gerd. and Trappe. Since N fertilization is of vital importance to quality of turfgrass, it would be worthwhile to determine whether mycorrhizae do in fact influence the downward movement of N in crushed basalt putting greens.

In view of the intense N management of golf greens and turf in general, the goal of this investigation was to determine the following:

1) the effect of two sources and three rates of N on nitrogen movement through turf grown on crushed basalt.

2) the effect of mycorrhizal infection on N movement through turf grown on crushed basalt.

3) responses of common bermudagrass in respect to visual rating, chlorophyll content, clipping dry weight, verdure dry weight, root dry weight, tissue analysis, and percent mycorrhizal infection to N source, N rate and mycorrhizal infection.

CHAPTER II

LITERATURE REVIEW

MYCORRHIZAE

The symbiotic relationship between vesicular-arbuscular mycorrhizal fungi and the roots of higher plants has been of interest to scientists for more than 100 years. Since growth response of mycorrhizal plants has been shown to be as much as 25 to 30 times greater than non-mycorrhizal plants (Kleinschmidt and Gerdemann, 1972), the enhancing effect of the fungi, due in part to increasing inorganic nutrient uptake, shows great promise as fertilizer sources become costlier as well as scarcer.

Mosse (1957) showed that the increase in growth of mycorrhizal plants was a reflection of improved plant nutrition. Studies similar to Mosse's comparing mineral composition of VA mycorrhizal plants have shown that phosphorus (P) is consistently higher in mycorrhizal plants (Gerdemann, 1975; Tinker, 1975).

Tinker (1975) summarized the hypotheses for mechanisms of increased P uptake by mycorrhizal plants as follows: (1) mycorrhizal infection alters root morphology perhaps by inducing hypertrophy in the roots, resulting in a larger root system and increased surface area for P absorption (2) mycorrhiza may have access to insoluble P sources such as apatite and tri-calcium phosphate (3) infection may alter host metabolism in such a way as to enhance the P absorption power of the host roots (4) increased soil volume utilized in nutrient absorption by hyphal uptake and subsequent translocation to the host roots (5) extended root activity time due to mycorrhizal infection. Hypothesis 4 seems to be the most credible as there is direct evidence that hyphal translocation of nutrients does occur (Cress et. al., 1979). The mechanism by which nutrients are translocated through hyphae to the host root appears to be one of cytoplasmic streaming (Cooper and Tinker, 1978; Cox et al., 1980). Increased absorption of other nutrients due to mycorrhizal infection have been demonstrated. Gilmore (1971) and La Rue et al. (1975) showed that zinc deficiency in peach (Prunus persica) was related to the absence of mycorrhizal infection. Furthermore, La Rue et al. (1975) demonstrated that subsequent mycorrhizal inoculation enhanced zinc uptake and improved growth more than additional zinc fertilization. Cooper and Tinker (1978) demonstrated the ability of Glomus mosseae to translocate zinc to host plants.

Mycorrhizal infection has also been shown to increase sulfur (S) uptake (Gray and Gerdemann, 1973) and sulfur can also be translocated to roots through external hyphae (Cooper and Tinker, 1978; Rhodes and Gerdemann, 1978).

Mycorrhizae are also involved in nitrogen (N) nutrition of legumes. Since NO_3, the N form primarily absorbed by plant roots, is very mobile in the soil and moves readily with the soil solution it stands to reason that the rate of movement to the root surface would not be a limiting factor in N uptake. Hyphal translocation of N therefore would be of little significance in plant N nutrition. Higher N concentration in leaf tissues as well as increased growth and improved yield in mycorrhizal-inoculated soybeans suggests that infection and subsequent nodulation enhances N uptake (Ross and Harper, 1970). Mycorrhiza and P fertilization had similar effects in increasing nodulation (Crush,1974; Daft and El-Giahmi, 1974). Either treatment also enhanced nitrogenase and nitrogen reductase activity (Daft and El-Giahmi, 1974; Carling et al, 1978) which would account for greater N concentrations in leaf tissues. Evidence that mycorrhizal infection does not increase N concentrations in non-legumes nor increase growth in non-nodulating isolines of soybeans grown under nitrogen deficient conditions (Schenk and Hinson, 1973; Carling et al, 1978) is evidence that improved N nutrition of legumes is not a direct effect of hyphal translocation but rather a secondary effect of increased N fixation due to greater P supply to roots.

Improved water availability to roots through a decrease in resistance to water transport as a result of mycorrhizal infection (Safir et al, 1971) apparently brought about by an improvement in plant nutrition is important. However, hyphal translocation of water under certain circumstances may be significant as well.

It had been assumed that nutrients were transferred from fungus to the host through the breakdown of arbuscules. However Cox and Tinker (1976) estimated that only a minute amount of P inflow to the host could be accounted for in this manner. A more likely mechanism of transfer across membranes was suggested. It has yet to be demonstrated that arbuscules are more efficient in transferring nutrients to the host root than undifferentiated hyphae or vesicles. Studies suggest that nutrient transfer from vesicles warrants further investigation (Cox and Tinker, 1976; Bowen et al., 1975).

NITROGEN

In light of costs for nitrogen fertilizers and the high N requirements of good quality turfgrasses, the rate at which N is leached out of the root zone is of utmost importance to the turfgrass manager. Loss of nitrogen for plant nutrition is dependent on several factors including the form of nitrogen applied, the porosity and infiltration rate of the soil and the management practices associated with the crop to be grown.

The two most common sources of nitrogen, ammonium and nitrate, behave quite differently under high leaching conditions. Availability to plants is very dependent on soil types. In soils containing large amounts of micaceous clay minerals such as vermiculite and montmorillinite, added ammonium ions are readily fixed (Nommik, 1957; Chaminade, 1971; Nielsen, 1972; Black and Waring, 1972; Raju and Mukhopadhyay, 1974). The mechanism of fixation of added ammonium appears to be the replacement of calcium, potassium, magnesium and sodium cations with ammonium ions in the expanded clay lattices. Timing of potassium applications influence the degree of ammonium fixation; K applications 7-10 days before ammonium fertilization minimizes NH_4^+ fixation (Sen Gupta et al., 1971; Raju and Mukhopadhyay, 1974). Although ammonium appears to be fixed preferentially, fixation of potassium ions on the expanded clay lattices prior to adding ammonium eliminates the fixing sites (Nommik, 1957; Sippola et al., 1973; Raju and Mukhopadhyay, 1973, 1974, 1976).

Nitrate nitrogen is not fixed in soils, however there are two pathways by which added NO_3^- becomes unavailable for plant nutrition. N may be lost through biological or chemo-denitrification (depending on soil pH) under reducing conditions. In fact, the presence of plants may enhance the denitrification process either by enlarging the anaerobic volume of the soil through restricted oxygen diffusion thereby increasing reducing conditions (Stefanson, 1972), or by providing nutrition to denitrifying microflora in the form of root exudates. Leaching of nitrate through the soil is by far the most common pathway of nitrate loss. The overall net negative charge of most temperate soils actually repels the NO_3^- ions, facilitating rapid movement of nitrate through the soil and out of the root zone. Some tropical soils containing amorphous inorganic compounds such as aluminum oxides and silicates tend to adsorb NO_3^- depending on pH and concentration of NO_3^- applied (Kinjo and Pratt, 1971a, 1971b; Kinjo Pratt and Page, 1971; Espinoza, Gast and Adams, 1975). This process by which nitrate is held in the root zone during the rainy season and during drier periods make nitrate leaching losses in tropical soils far less than in temperate soils.

Nitrate absorption is an active, energy-requiring process. The rates of absorption of ions from liquid culture by most higher plants remain relatively constant during the initial stages of ion uptake. On the other hand, nitrate absorption by nitrate-depleted plants has been shown to exhibit an early lag period followed by a more rapid rate of absorption. This apparent induction period has been observed in wheat (Minotti, Williams and Jackson, 1969b) and many other crops. The accelerated rate after induction seems to be dependant on a critical internal nitrate concentration. This critical level is a function of the external concentration (Neyra and Hageman, 1975). It has also been shown that under normal conditions, nitrate uptake and nitrate-reductase activity are closely correlated. Neyra and Hageman (1975) suggest the reason for this may be that the rate of nitrate-reductase induction is regulated by the nitrate flux into the plant.

Ammonium inhibition of uptake and reduction of nitrate has been found in wheat (Minotti et al., 1969a) as well as other crops. Minotti et al. (1969b) suggest the reason for this may be that ammonium, and to some extent the high acidity adjacent to the cellular boundary membranes created by more ammonium being taken up than nitrate, caused the membrane permeability to become altered which restricts the capacity for nitrate absorption.

Plants grown in either ammonium or nitrate solutions change the pH of their medium with time. Blair et al., (1970) and others have shown that the media containing nitrate becomes more alkaline and those with ammonium become more acid. This is true for a number of plant species. This change in pH seems to be due to excretion of hydrogen ions by the roots when ammonium is taken up and hydroxyl ions when nitrate is taken up. The exact mechanism is complicated but results point to a complex hormone-regulated mechanism.

It has been shown that low temperatures inhibit nitrate uptake more than ammonium uptake (Williams and Vlamis, 1962). They also found that in nutrient solutions, nitrate absorption was affected more by lower temperatures than other ions.

There is a relationship between carbohydrate supply and ammonium and nitrate uptake (Michael et al., 1970). When ammonium is absorbed, it can be used to synthesize organic molecules. This can be thought of as a detoxification process since high levels of ammonium in the plant do become toxic. At these high levels, depletion of carbohydrates due to synthesis of amino acids and amides will occur so carbohydrate supply is of some importance in ammonium nutrition. Nitrate is not as toxic as ammonium and may be stored in vacuoles of the roots or translocated to the shoots and must be reduced before it can be used, so it is less affected by carbohydrate supply.

It has been noted that plants grown in ammonium medium without nitrate in general contain lower concentrations of calcium, magnesium and potassium and higher levels of phosphorus and sulfur than those supplied with nitrate alone. Cox and Reisenauer (1973) using wheat as their indicator plant showed that as nitrate uptake rates increased, so did calcium, magnesium, and potassium but with increasing rates of ammonium absorption, intake of calcium and magnesium decreased. These higher rates of magnesium calium and potassium intake with nitrate as opposed to ammonium can be attributed to reduced competition in the absorption process.

It has also been shown that when ammonium replaced nitrate as the N source, phosphorus and sulfur uptake increased. When ammonium N was applied during the fruiting stage of tomato, there was a rapid development of blossom-end rot of the fruit due to the inhibition of calcium uptake by ammonium (Wilcox et al., 1973).

It has been assumed that most plants take up most of their nitrogen in the nitrate form. However, there are some plants, such as some species of Pinus, which prefer ammonium as their N source and will grow in nitrate deficient soils. These types of plants show no calcium or magnesium deficiency symptoms when subjected to ammonium nutrition. The physiological reason for this is still unknown.

Comparisons between a controlled release fertilizer ("Osmocote") and a continuous liquid feed fertilizer show that a controlled release fertilizer is much more efficient in reducing nitrogen loss via leaching and increasing N recovery than a continuous liquid feed fertilizer when surface irrigation is applied (Hershey and Paul, 1982). The nitrogen lost was expressed as a percentage of nitrogen applied and in both the controlled release fertilizer and the continuous liquid feed, more nitrogen was lost where greater amounts of nitrogen were applied. Previous studies (Holcomb, 1979; Barron, 1977) indicate that controlled release fertilizer applications are much more efficient and therefore economical in the cultivation of potted chrysanthemums.

The particle size, shape and distribution of sand used in soil mixes for growing high quality turfgrasses affects the porosity and water infiltration rate (Duble and Brown, 1975) and therefore the leaching rate. Porosity, or the lack thereof, under intense traffic conditions associated with golf putting greens can affect denitrifying processes by decreasing the aeration pore space and thereby producing anaerobic conditions conducive to denitrifying microflora (Stefanson, 1972). However, a golf course putting green built to USGA Greens Section specifications will not usually experience problems with denitrification because the soil mix is made specifically to withstand compaction and maintain sufficient aeration porosity to avoid reducing conditions in the root zone. By the same token, the mix provides for rapid infiltration rates (Brown and Duble, 1975) which increases the rate at which nitrate fertilizers are carried downward through the soil and out of the root zone. Intense N fertilization of golf greens with soluble sources increases the amount of NO_3^- leached (Hershey and Paul, 1982) from soil mixes with high infiltration rates and from run-off in mixes with lower infiltration rates.

In addition to the obvious economic reasons to minimize leaching losses of NO_3^- from fertilizer application, the further repurcussions of excess energy consumption to produce more N fertilizer and the possibility of NO_3^- contamination of ground water used for drinking must be considered (Starr and DeRoo, 1981).

Definitive data on the influence of management practices on nitrogen losses through leaching are few (Brown, Duble and Thomas, 1977). However techniques to minimize NO_3^- leaching by regulating the amount and timing of irrigation applied have been investigated. La Rue et al. (1968) and Tovey (1969) found that heavy infrequent irrigations lessened nitrate losses as opposed to frequent, lighter waterings.

CHAPTER III

MATERIALS AND METHODS

Simulated golf putting greens were constructed to study the influence of nitrogen source and rate of application on nitrogen leaching under common bermudagrass (*Cynodon dactylon* L. Pers.) inoculated and uninoculated with a vesicular-arbuscular mycorrhizal (VAM) fungus (*Glomus mosseae* (Nicol. and Gerd.) Gerd. and Trappe).

Simulated putting greens were constructed in polyvinyl chloride (PVC) cylinders 15.2 cm diameter X 48.3 cm long. Each PVC cylinder was cemented onto an acrylic plastic base and fitted with 4 acrylic plastic drainage tubes of 0.9 cm inside diameter (Figure 1). Cemented cylinders were allowed to set for 24 hours, drainage tubes were stoppered, and cylinders were checked for leaks by filling with water. All leaks were sealed before the cylinders were used.

A 10.2 cm drainage layer of gravel sized (approx. 0.6 cm diameter) crushed basaltic rock was placed in the bottom of each cylinder after covering the outlet of the drainage tubes with plastic screen. A 5.1 cm filter layer of coarse sand sized (1.0 mm - 2.0 mm) crushed basaltic rock was placed over the drainage layer. Finally, a 30.5 cm root zone layer of coarse to medium sized (95% by weight in the 0.25 - 1.0 mm range) crushed basaltic rock was placed over the filter layer. This construction is similar to

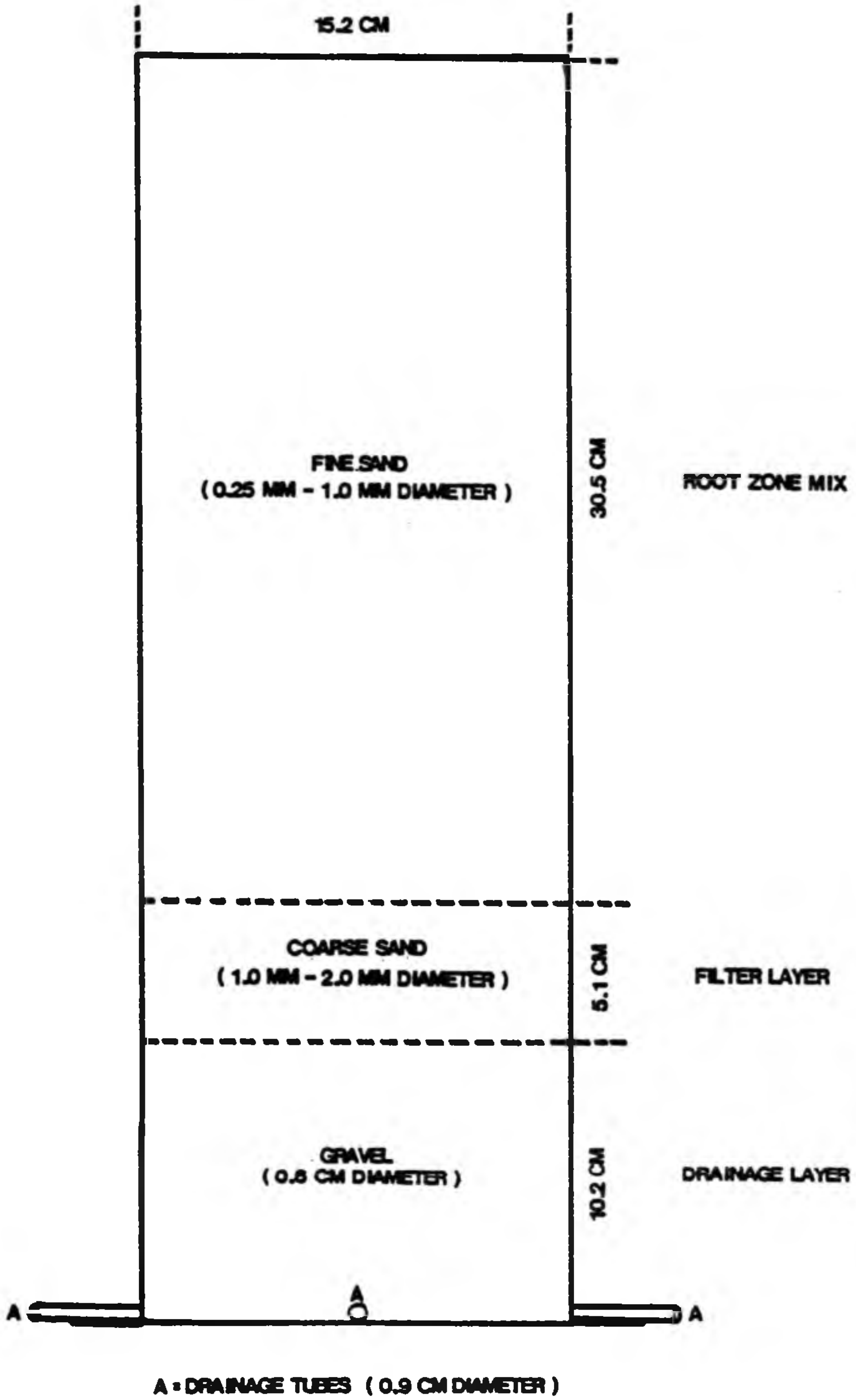

FIGURE 1. SCHEMATIC DIAGRAM OF SIMULATED GOLF PUTTING GREEN

procedures recommended by the U.S. Golf Association Green Section (Ferguson 1965). Crushed basaltic rock materials were used in constructing these simulated greens, as this is the only locally available material presently used by turf managers in Hawaii for amending high-traffic soils.

The PVC cylinders were surface sterilized with 50% methanol. Basaltic rock materials were irradiated with a Co^{60} source (2Mrad for 6 hours). One-half of the cylinders were inoculated with *Glomus mosseae* and one-half were uninoculated. The *G. mosseae* culture was originally obtained from Dr. J.W. Gerdemann, University of Illinois. It was increased on corn (*Zea mays* L.) cv. "Hawaiian Supersweet #9" in basaltic sand. Examination of cultures after increasing the inoculum revealed a small number (less than 5% of total) of spores of *G. etunicatus* Becker and Gerd. It is not known if the original culture was contaminated or if contamination occurred in our cultures. Because of the small amount of contamination, the culture was used in this research.

Spores were obtained by wet sieving and decanting (Gerdemann and Nicholson, 1963) sand and roots from the pot cultures. Approximately 100 spores were distributed evenly over the surface of partially filled cylinders (approximately 10 cm below the final surface). A filtrate obtained by washing spores with deionized water and filtering through Whatman No. 1 paper was added to uninoculated cylinders to assure addition of contaminating fungi and bacteria. After inoculum was added, cylinders were completely filled with sand, tamped lightly to settle, and seeded with common bermudagrass. Cylinders were placed under mist until germination occurred.

Fourteen days after turfgrass germination, cylinders were moved to a glasshouse bench and given 250 ml of a modified Hoaglands No. II (Hoagland and Arnon, 1951) solution containing 25% of the normal $NH_4H_2PO_4$ level plus micronutrient solution. This solution was applied 3 times per week on alternate days. In addition, a 0.5% iron citrate solution was applied once a week. Simulated greens were leached with 1 liter deionized water weekly to prevent salt accumulation. Grass was trimmed at 5.1 cm height every other week and sprayed as needed with a 0.2% Chloropyrifos solution to control Rhodesgrass scale (*Antonina graminis* Maskell).

Root samples were taken at 2, 6 and 10 months after grass was established by inserting a 1 cm diameter cork borer to a depth of 6 cm in each root zone. Roots were washed free of sand, cleared and stained (Phillips and Hayman, 1970) and examined for VAM infection. Simulated golf greens were maintained in this manner for approximately one year before applying nitrogen treatments. Final examination of roots showed a high degree of VAM infection in all inoculated cylinders and none in uninoculated ones.

A factorial treatment set consisting of two VAM levels (inoculated and uninoculated), two nitrogen sources, ($Ca(NO_3)_2$ and $(NH_4)_2SO_4$), and three levels of N application (24.4, 48.8 and 73.2 kg/ha) was replicated three times in a completely random design.

Nitrogen treatments were applied at 14 day intervals in 100 ml deionized water distributed evenly over the turf surface. Cylinders were leached by adding 1 liter (5.5 cm over the surface) deionized water to each cylinder the day after adding N treatments and every other day thereafter. It had previously been determined that addition of 1 liter of water resulted in approximately 500 ml of water leaching through the cylinder. The nitrogen application and leaching schedule was repeated for 3 cycles over a 6 week period. A total of 21 leachings were obtained during this period.

Leachates were collected and measured at each leaching date. Total-N and NO_3-N content of each leachate was determined. An Orion nitrate specific ion electrode attached to an Altex model 4500 digital pH meter was used to determine NO_3^- concentration. The "known addition" technique was employed (Orion Research Inc., 1973) as NO_3^- concentration of many of the leachates was below the sensitivity level of the electrode. Total-N was determined by a modification of the method described by Cataldo et al. (1974). Twenty-five ml of leachate was substituted for 50 g of plant tissue in the analysis.

Turf was clipped every other week throughout the experiment to a height of 5.1 cm. Clipping dry weights were recorded after drying for 48 hours at 60 degrees C in a forced air dryer. Chlorophyll content was obtained from the dried tissue by modification of the method described by Johnson (1974). Chlorophyll was extracted from twenty mg of dried ground clippings with 20 ml of methanol for 24 hours. Optical density (OD) at 650 and 665 nm was determined with a Unicam SP 1800 Ultraviolet Spectrophotometer. Chlorophyll content was calculated by the formula $(25.6 \times OD_{650} + 4.0 \times OD_{665})$ and expressed as mg of chlorophyll/g clippings.

Visual ratings of turf quality based on color, density and uniformity were made on a scale of 1 to 5 with 1 being poorest and 5 best quality. A rating of 3 was considered acceptable turf. All ratings were made at approximately 7:30 A.M. to standardize lighting conditions. Ratings were obtained every other week at the end of each cycle.

Verdure (Madison, 1962) dry weights were measured at the end of the experiment. After clippings were obtained for the last time, plant material remaining was harvested to sand level, dried weighed and recorded.

Root dry weights were measured by washing roots free of sand and drying. Subsamples from three depths (5.5 to 9.0 cm, 21.5 to 25.5 cm and 38.5 to 42.5 cm) were obtained for determining VAM infection. Fresh weight of these subsamples was recorded. A similarly sized sample from the same depths was taken, fresh and dry weights recorded, and dry weights of the sample used for VAM determination were calculated to obtain total root system dry weight.

Degree of VAM infection at the three depths of each root system was determined after clearing and staining (Phillips and Hayman, 1970) the root subsamples. Since bermudagrass roots are very fine and fibrous, it was necessary to use a compound microscope (100X) for estimating degree of VAM infection. Fifty fields of vision were inspected on subsamples from each root system from each depth. Each field of vision was recorded as infected if any VAM structure (vesicles, arbuscules, hyphae or chlamydospores) were present in the roots. Percent infection was calculated by dividing the number of fields of vision with infected roots by 50 and multiplying by 100.

Tissue analysis was conducted (on dry clippings and roots) for percent N, P, K, Ca, Mg, S, Si, and ppm Al, Mn, Fe, Zn and Cu by X-ray quantometry, Soil and Tissue Analysis Laboratory, College of Tropical Agriculture and Human Resources, University of Hawaii. Root samples for tissue analysis were taken from 18-28 cm depth below the surface.

Analysis of variance (ANOVA) was performed on all data. The HP 2000 computer, University of Hawaii Computing Center, was used in statistical analysis. Each 14 day cycle (and subsequent 7 leachings) was analyzed and plotted separately.

Bayesian LSD (Waller and Duncan, 1969) at the 0.05 level of probability was used to compare differences between treatment means.

CHAPTER IV

RESULTS AND DISCUSSION

I. VAM INFECTION

Mycorrhizal infection was very obvious in all inoculated pots as indicated by vesicles, spores and arbuscules. However, there were not many hyphae present. This was thought to be due to loss of hyphae when washing roots or to an inactive or slow stage of fungal growth at the time of root harvest.

With increasing depths, there was a significant decrease (Anova, $p<.01$) in percent VAM infection. Each depth had a significantly lower infection level than the preceeding depth (Table 1). This result has been shown to occur with different grasses (Bagyaraj et al., 1980). Their results showed that percent infection with a VAM fungus was significantly higher at 3 - 4 cm depth than 8 - 9 cm except for one grass. They also found that the overall percent infection in different grasses varied upholding the view (Mosse, 1977) that plant species differ in their susceptibility to mycorrhizal infection.

It was also observed that a significantly higher (Anova, $p<.05$) percent infection level of the whole root system occurred when ammonium sulphate was the source of N (Table 2). It is known that most fungi prefer ammonium-N

TABLE 1. EFFECT OF DEPTH ON PERCENT VAM INFECTION IN ROOTS OF COMMON BERMUDAGRASS AT THE FINAL HARVEST[w]

Depth (cm)	Percent Infection
5.5-9.0	68a[*]
21.5-25.5	48b
38.5-42.5	30c

[*]Means for treatment effects followed by the same letter differ significantly (BLSD=.05).

[w]Average of 2 N sources and 3 N levels.

TABLE 2. EFFECT OF N SOURCE ON PERCENT VAM INFECTION AT THREE SAMPLING DEPTHS IN ROOTS OF COMMON BERMUDAGRASS AT THE FINAL HARVEST[w]

	Percent Infection			
	Depth (cm)			
N Source	5.5-9.0	21.5-25.5	38.5-42.5	Average
$Ca(NO_3)_2$	61.3	41.9	28.4	44.4*
$(NH_4)_2SO_4$	75.8	54.4	32.0	54.1

*Means in the same column are significantly different ($p=0.05$).

[w]Average of 3 N levels.

(Burnett, 1968) and this form is apparently a major source of N for ectomycorrhizal fungi (Carrodus, 1966, 1967; Lundeberg, 1970). However nothing conclusive can be said since nitrate leached much more readily than ammonium and since soil N levels were not monitored.

It is well known that high levels of N as well as P reduce mycorrhizal infection (Lanowska, 1966; Hayman, 1970). This was noted to occur with ammonium sulphate (Table 3) where infection levels decreased with increasing levels of N but was not noticed with calcium nitrate possibly because of the high leaching rates in the experiment moving most of the N out of the root zone.

II. NITROGEN LEACHING

NO_3-N and total-N content of leachates were measured on alternate days for a period of 2 weeks after treatments were applied. The treatment and leaching schedule was repeated for three 2 week cycles. Twenty-one leachings were obtained during this period.

A. NITRATE

In all cases, there was a significant effect of time (Anova, $p<.01$) on NO_3-N present in the leachate (Table 4). In general, most of the NO_3-N was lost during the first six days after N application, with no differences between means

TABLE 3. EFFECT OF N SOURCE AND N LEVEL ON PERCENT VAM INFECTION AT THREE SAMPLING DEPTHS IN ROOTS OF COMMON BERMUDAGRASS AT THE FINAL HARVEST

		Percent Infection			
		Depth (cm)			
N Source	N Level (kg/ha)	5.5-9.0	21.5-25.5	38.5-42.5	Average
$Ca(NO_3)_2$	24.4	58.7	42.7	26.7	42.7
	48.8	60.3	43.3	32.7	45.4
	73.2	66.3	39.7	26.0	44.0
$(NH_4)_2SO_4$	24.4	90.7	52.7	40.0	61.1
	48.8	68.0	56.7	32.7	52.4
	73.2	68.7	54.0	23.3	48.7

TABLE 4. EFFECT OF TIME IN DAYS ON NITRATE-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Nitrate-N (kg/ha)		
Day	Cycle 1	Cycle 2	Cycle 3
2	2.63a*	3.45a	2.69b
4	1.97ab	2.96a	3.73a
6	1.43bc	1.54b	1.81c
8	0.88c	0.77c	0.71d
10	0.82c	0.66c	0.66d
12	0.71c	0.66c	0.60d
14	0.77c	0.60c	0.60d

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources, 3 N levels, 2 mycorrhizal levels.

TABLE 5. EFFECT OF N SOURCE ON NITRATE-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Nitrate-N (kg/ha/leaching)		
N Source	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	1.81*	2.19*	2.14*
$(NH_4)_2SO_4$	0.88	0.88	0.93

*Means in the same column are significantly different ($p=0.05$).

[w]Average of 3 N levels, 2 mycorrhizal levels and 7 leaching dates.

after this. In all cases, more N was leached from calcium nitrate (Anova, $p<.01$) in the nitrate form than from ammonium sulphate (Table 5). Greater amounts (Anova, $p<.01$) of NO_3-N were leached from the high N rate (Table 6) with no difference between the low and medium rates. Total NO_3-N leached at the end of each 2-week cycle was greater for calcium nitrate than ammonium sulphate (Figures 2, 3, 4). Generally, with increasing rates of calcium nitrate there were correspondingly increasing amounts of NO_3-N lost in the leachate. This trend also held true for ammonium sulphate but to a much lesser extent. It is well known that NO_3^- moves much more readily in the soil solution than does NH_4^+ under high leaching conditions, due to the fact that NH_4^+ is fixed on soil colloids while NO_3^- is not (Black and Waring, 1972; Nielsen, 1972). It therefore follows that the greater the NO_3^- concentration in the soil solution, the greater the nitrate leaching loss will be, all other things held constant. In general, with increasing rates of calcium nitrate total NO_3-N leached at the end of each 2 week cycle, expressed as a percent of N applied, increased and decreased with increasing rates of ammonium sulphate (Figures 5, 6, 7). The ammonium sulphate trend agrees with that found by Brown et al (1977). Although they did not include calcium nitrate as a treatment, their results for ammonium sulphate were 37.8%, 24.9% and 22.0% NO_3-N leached for the low,

TABLE 6. EFFECT OF N LEVEL ON NITRATE-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Nitrate-N (kg/ha/leaching)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3
24.4	0.88b*	0.77c	0.71b
48.8	1.10b	1.37b	1.21b
73.2	1.97a	2.47a	2.74a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources, 2 mycorrhizal levels and 7 leaching dates.

TABLE 7. EFFECT OF N SOURCE AND N LEVEL ON NITRATE-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Nitrate-N (kg/ha/leaching)		
N Source	N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	0.88b*	0.82c	0.71c
	48.8	1.37b	1.86b	1.54b
	73.2	3.13a	3.89a	4.11a
$(NH_4)_2SO_4$	24.4	0.82b	0.71c	0.66c
	48.8	0.82b	0.88c	0.88bc
	73.2	0.88b	0.99c	1.32bc

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 mycorrhizal levels and 7 leaching dates.

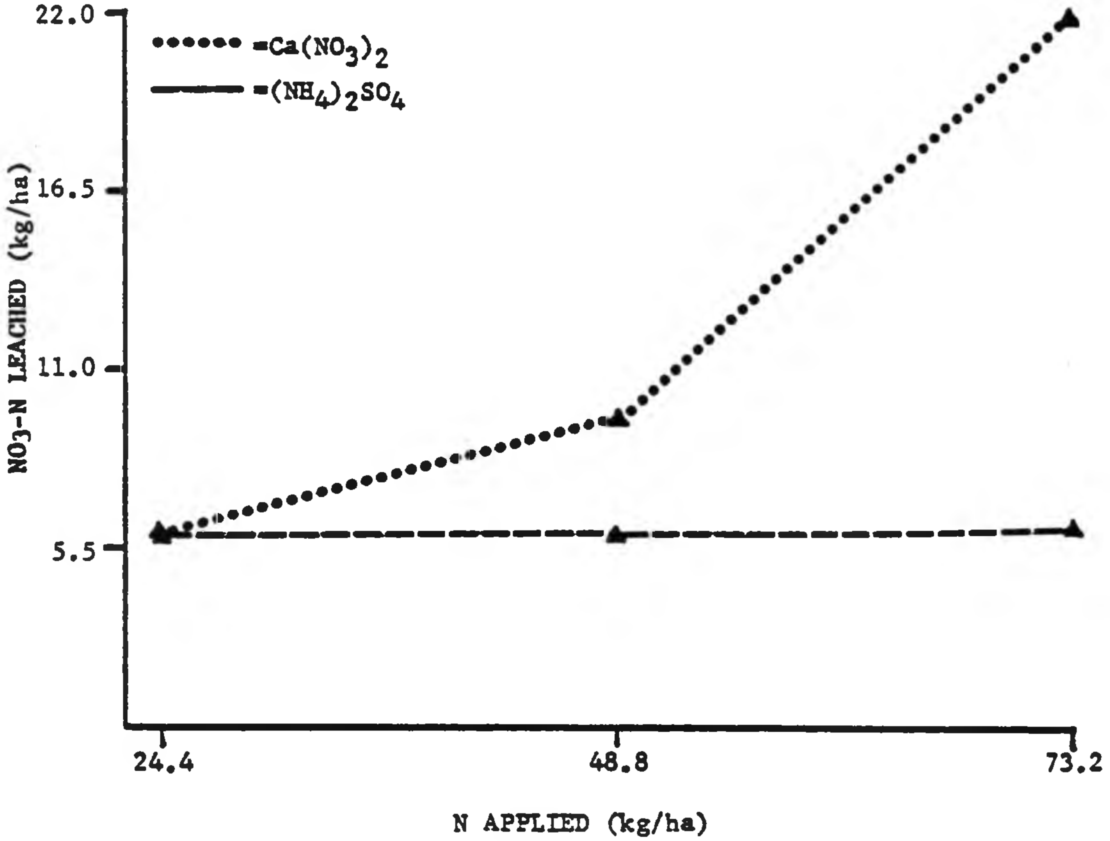

Figure 2. Nitrate-N leached with calcium nitrate and ammonium sulfate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the first 2-week cycle).

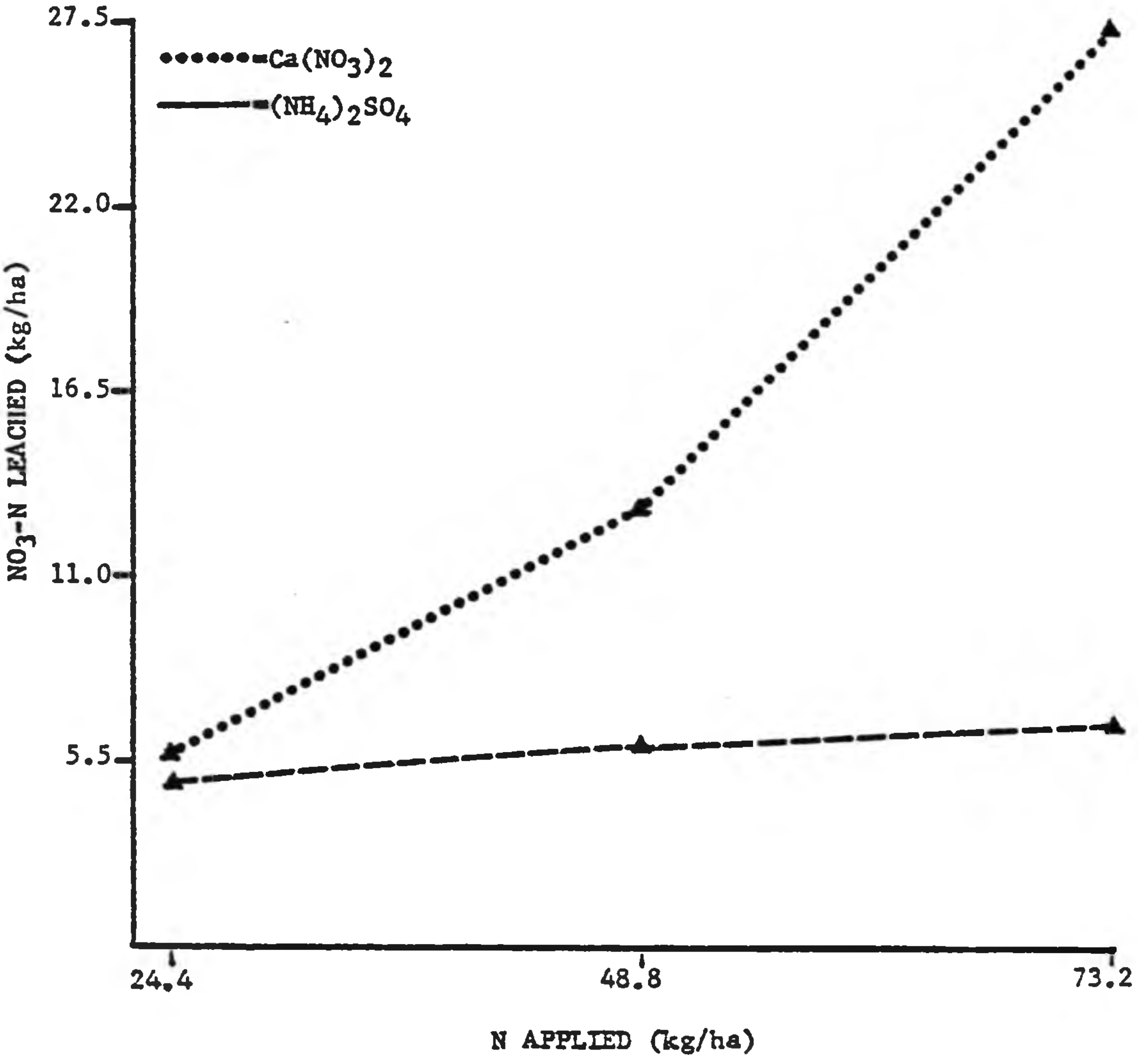

Figure 3. Nitrate-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the second 2-week cycle).

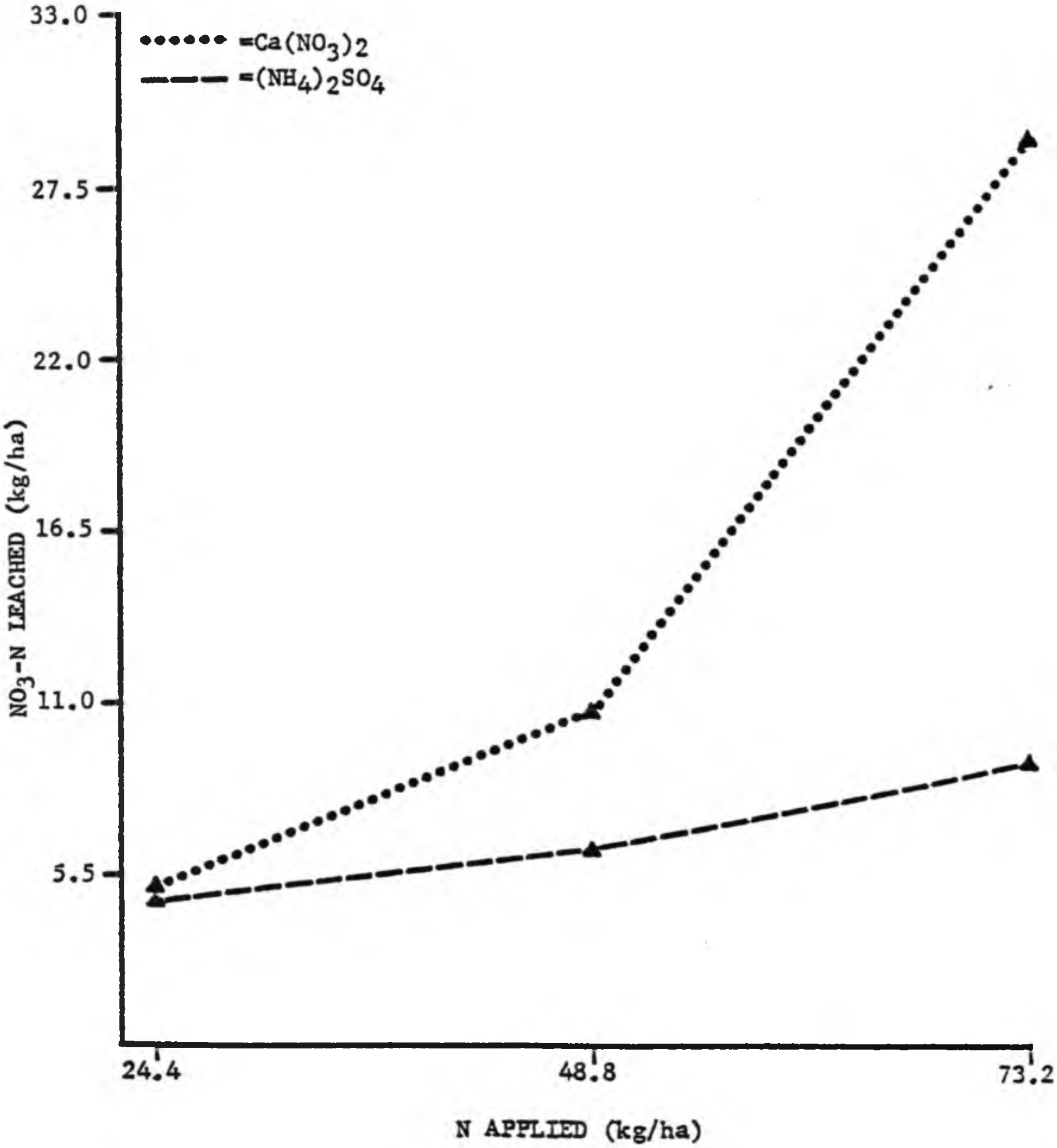

Figure 4. Nitrate-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the third 2-week cycle).

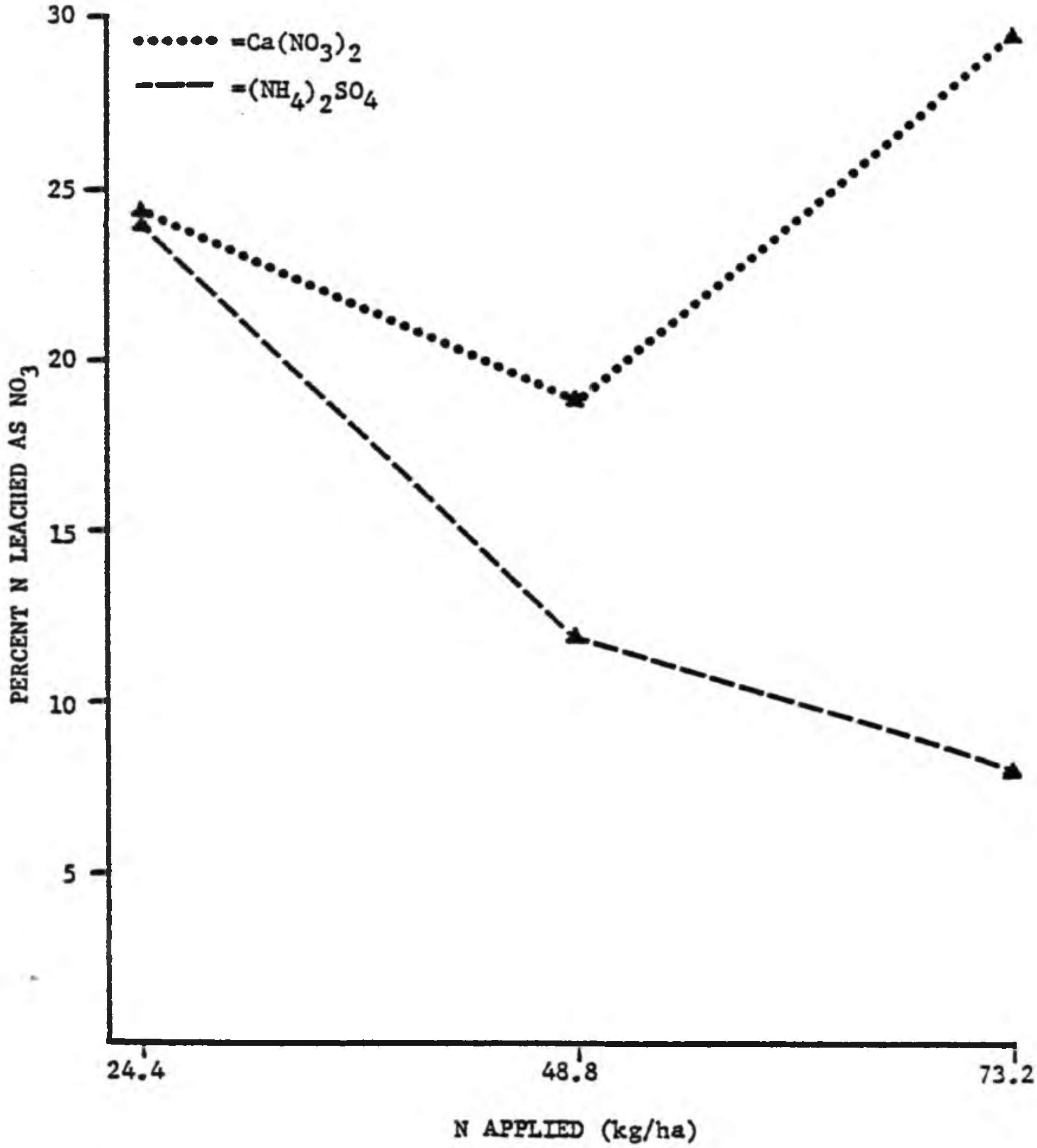

Figure 5. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the first 2-week cycle).

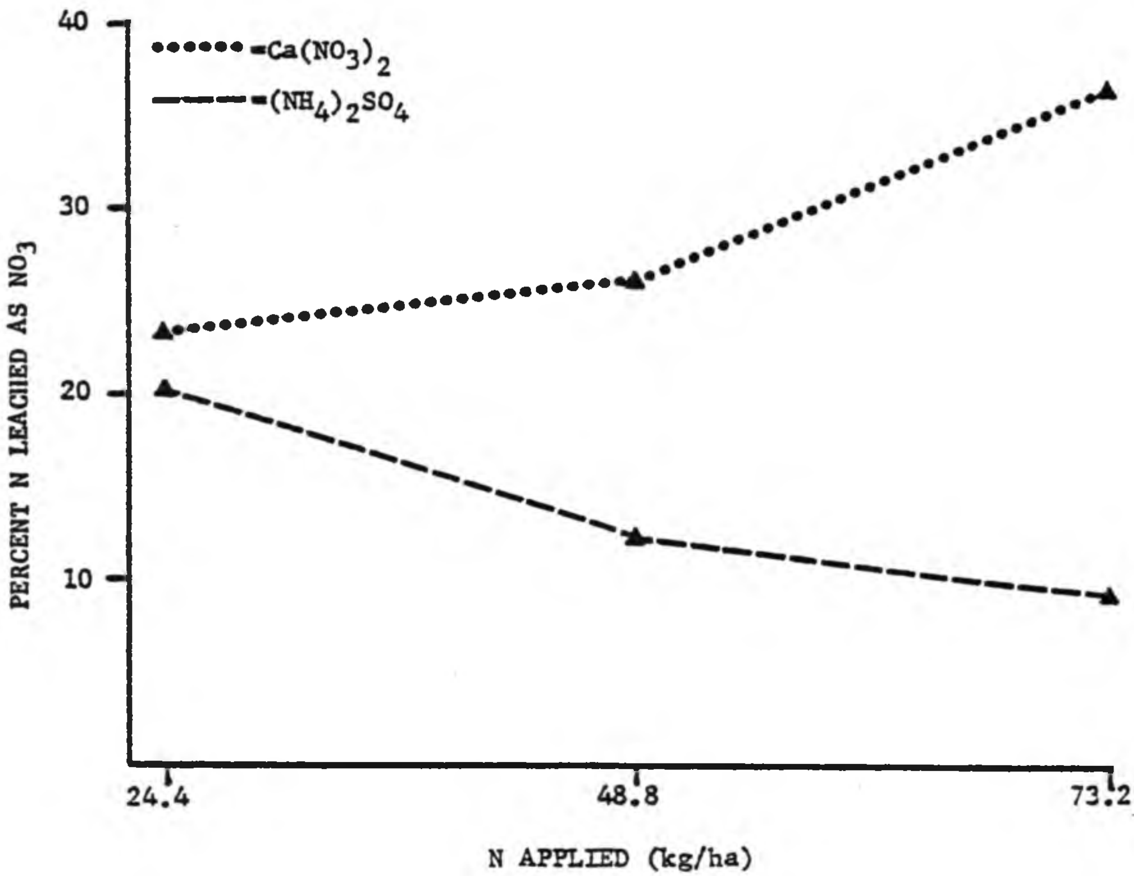

Figure 6. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the second 2-week cycle).

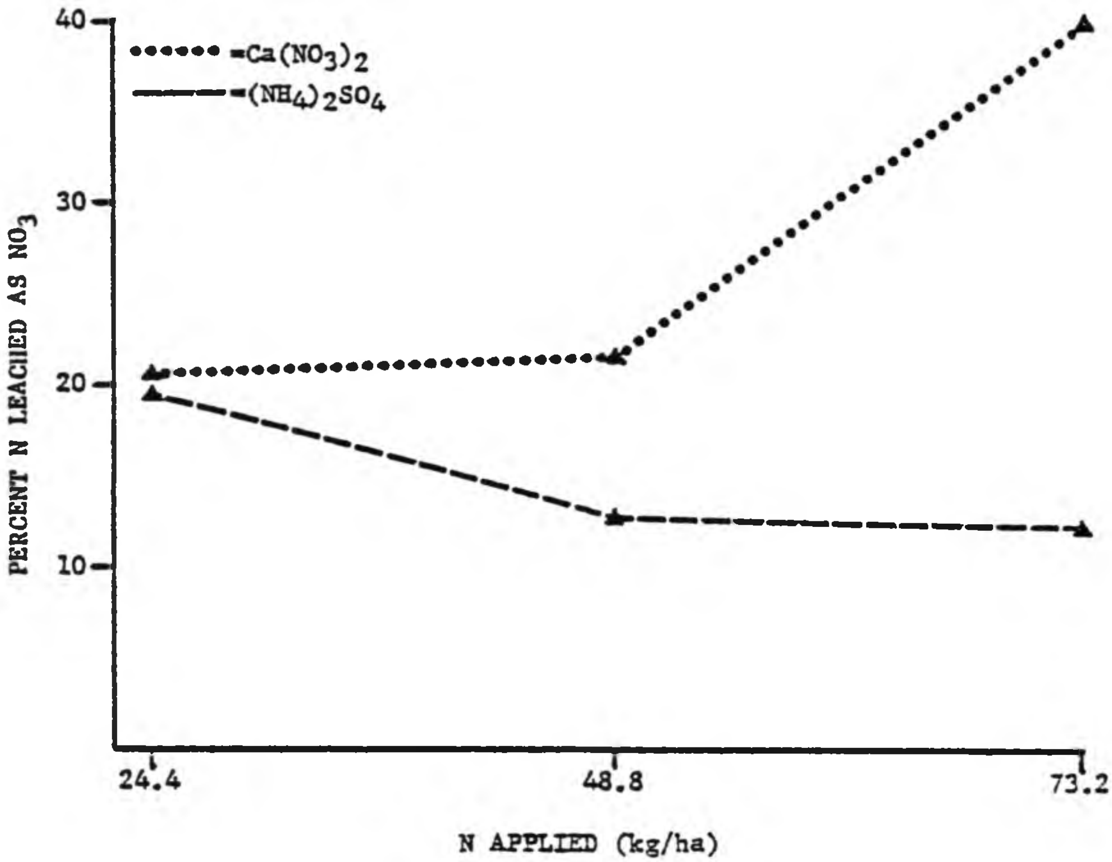

Figure 7. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the third 2-week cycle).

medium and high rates as compared to 21.2%, 12.3% and 9.8% (averages of 3 cycles) respectively in our experiments. The differences between percent N lost could be due to the fact that lower irrigation rates were used by Brown, allowing more time for nitrification and thus greater NO_3^- loss when irrigation was applied or rain occurred. The fact NO_3-N leached from ammonium sulphate as a percent of N decreased suggests that perhaps soil nitrifier populations remained fairly constant regardless of ammonium application rate. This may have been due to the rate of absorption of ammonium by the plant roots, which could limit the amount of substrate available to the nitrifiers.

A significant interaction (Anova, $p<.01$) occurred between N source and rate in respect to NO_3-N leached. Table 7 shows that there was essentially no difference in the amount of NO_3-N leached between all ammonium sulphate means and the low calcium nitrate. On a daily basis, NO_3-N leached was highest at the high calcium nitrate rate with leveling off occurring after the sixth day (Figures 8, 9, 10). The medium calcium nitrate rate was the next highest, with leveling off occurring after the fourth day. The three ammonium sulphate levels had about the same leaching rate as the low level of calcium nitrate in the first 2 week cycle (Figure 8). In succeeding cycles, however, more NO_3-N was

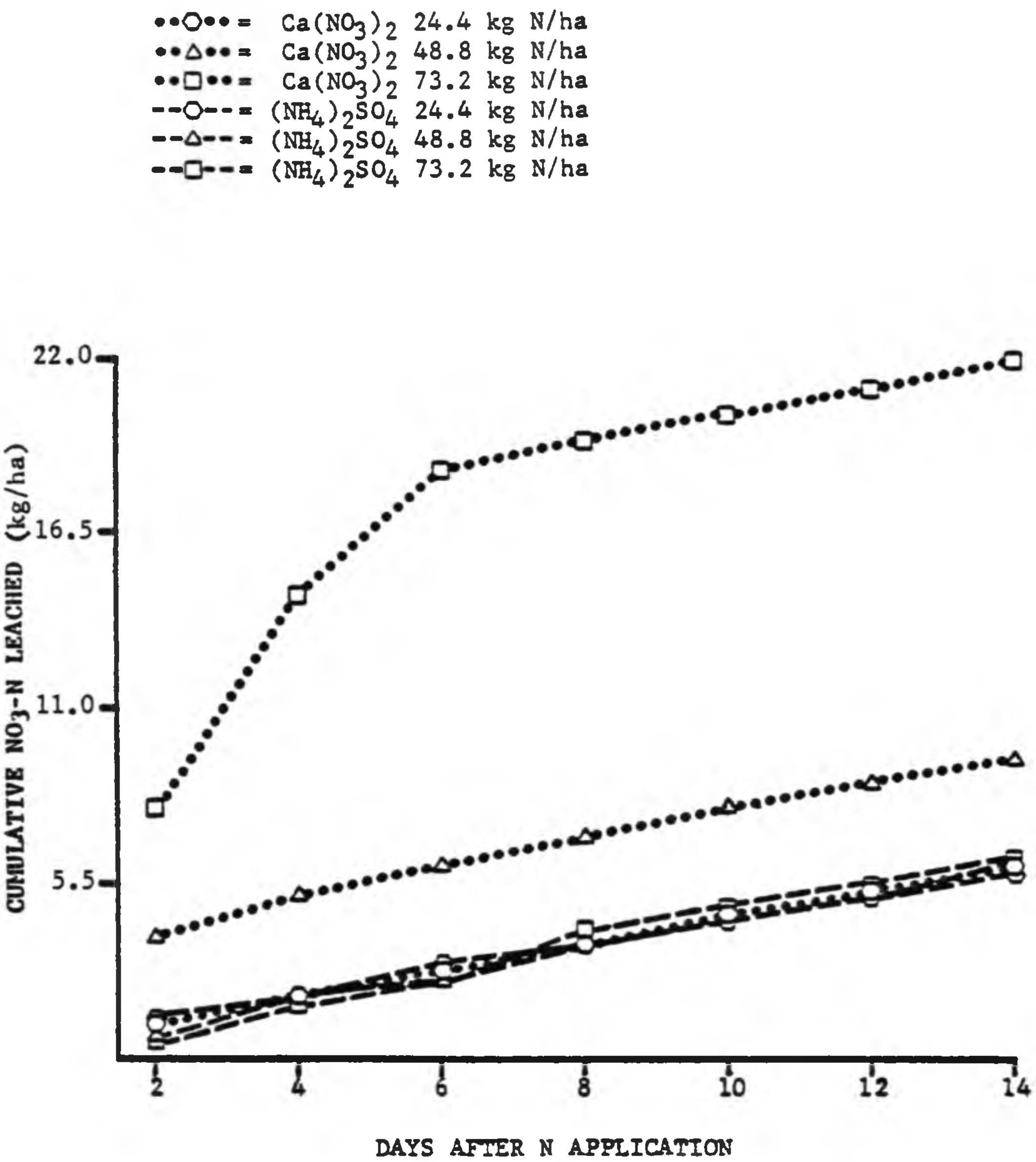

Figure 8. Nitrate-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the first 2-week cycle).

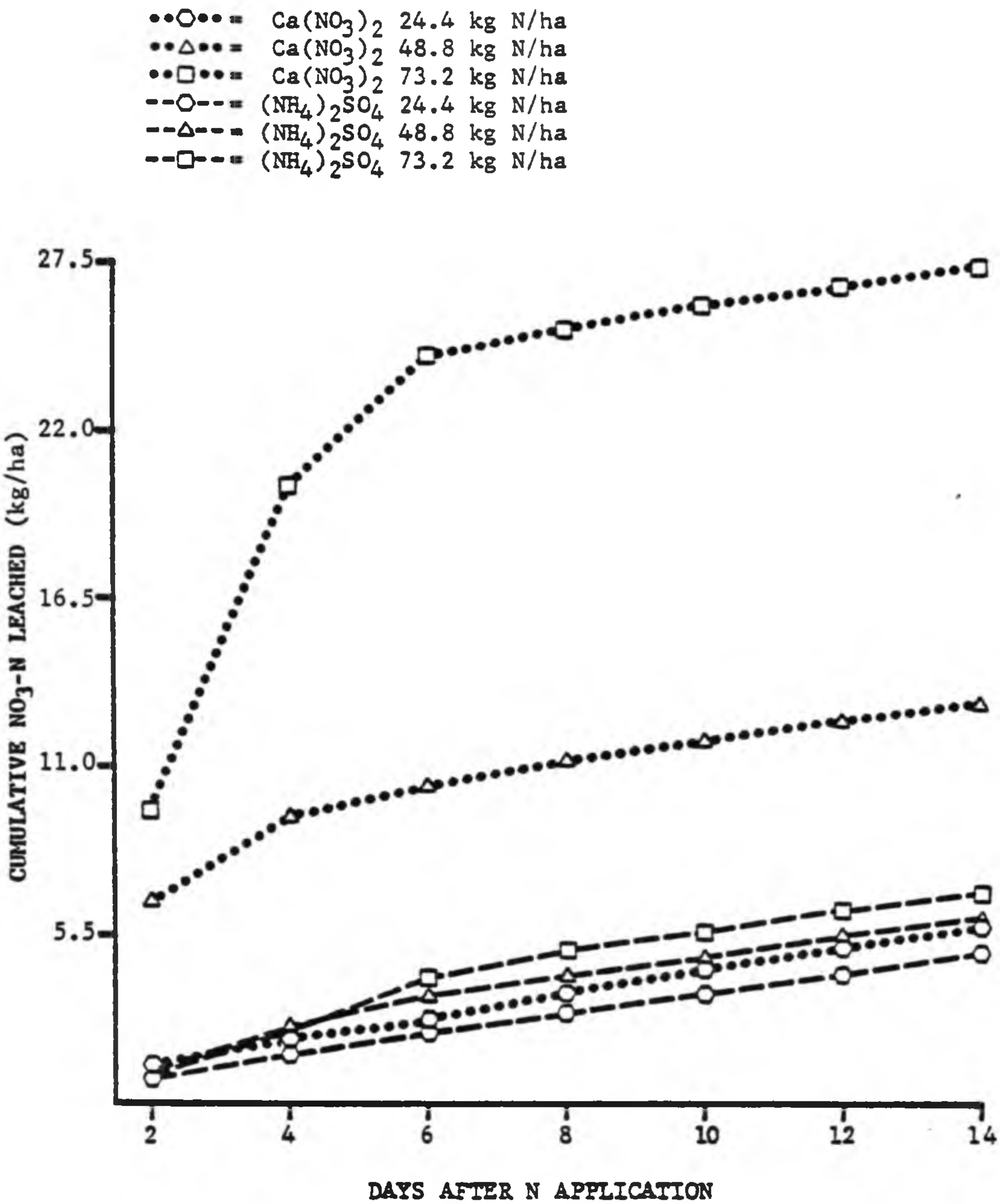

Figure 9. Nitrate-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the second 2-week cycle).

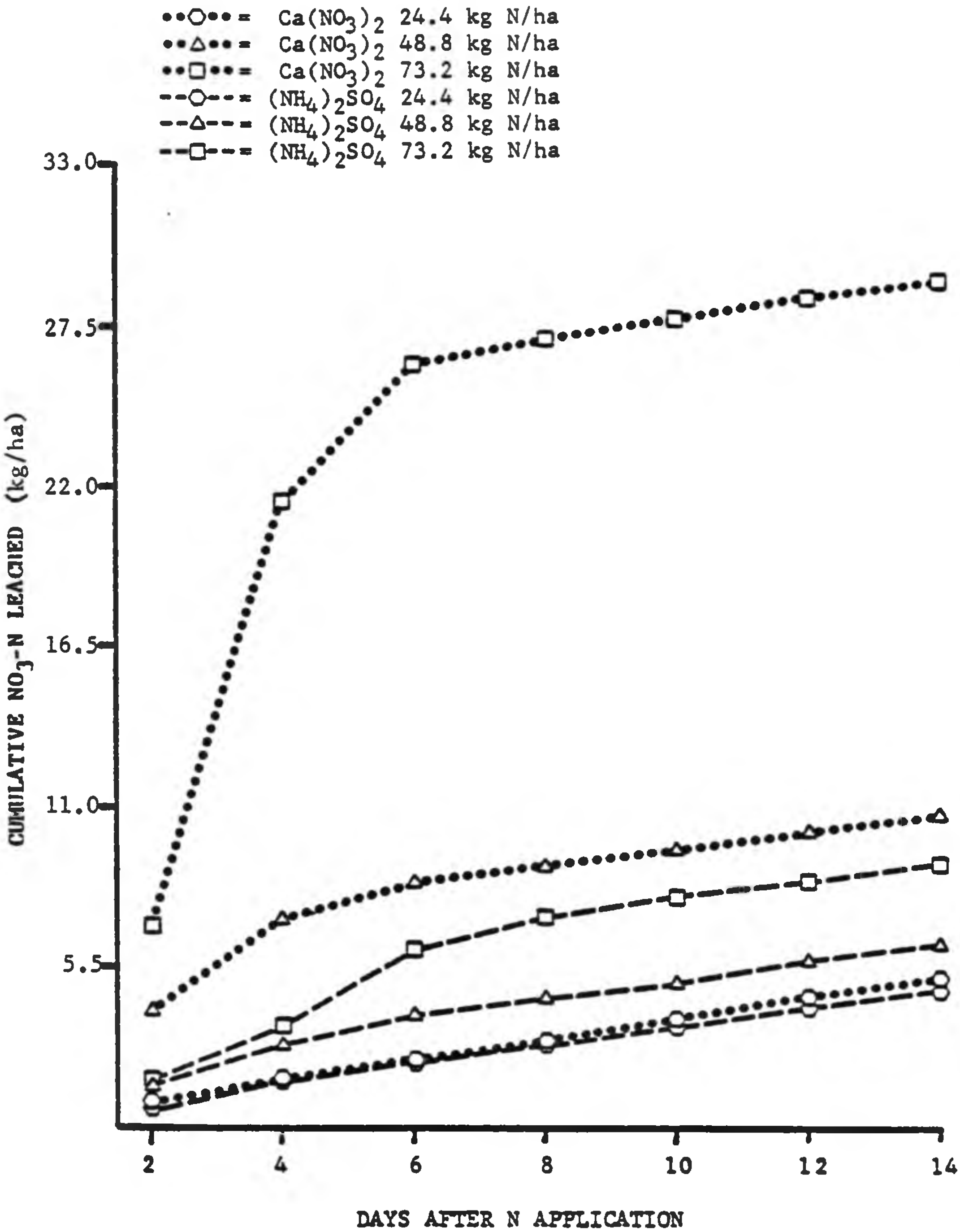

Figure 10. Nitrate-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the third 2-week cycle).

leached from the medium and high levels of ammonium sulphate (Figures 9 and 10). Since no N application was made for several months prior to the start of the experiment, nitrifier populations were probably low. Thus as substrate became available, populations grew and nitrification increased.

Figures 11, 12 and 13 express NO_3-N loss as a cumulative percent of N applied. Due to the high cost of N fertilizers, it is important to note that after the first leaching (day 2), about 10% of the N applied had already been lost with the high rate of calcium nitrate and from 25% - 35% by the sixth day whereas with the high rate of ammonium sulphate, only 2% was lost as NO_3-N after the first leaching and about 5% by the sixth day. Increasing rates of calcium nitrate had correspondingly increasing percentages of N leached while the opposite trend occurred with increasing rates of ammonium sulphate.

There were no observed mycorrhizal effects on NO_3-N leached.

B. TOTAL-N

When looking at values of N leached as total-N, it should be noted that at the low and some medium N fertilization rates, readings may be lower than NO_3-N readings indicating a loss of accuracy with one of the N

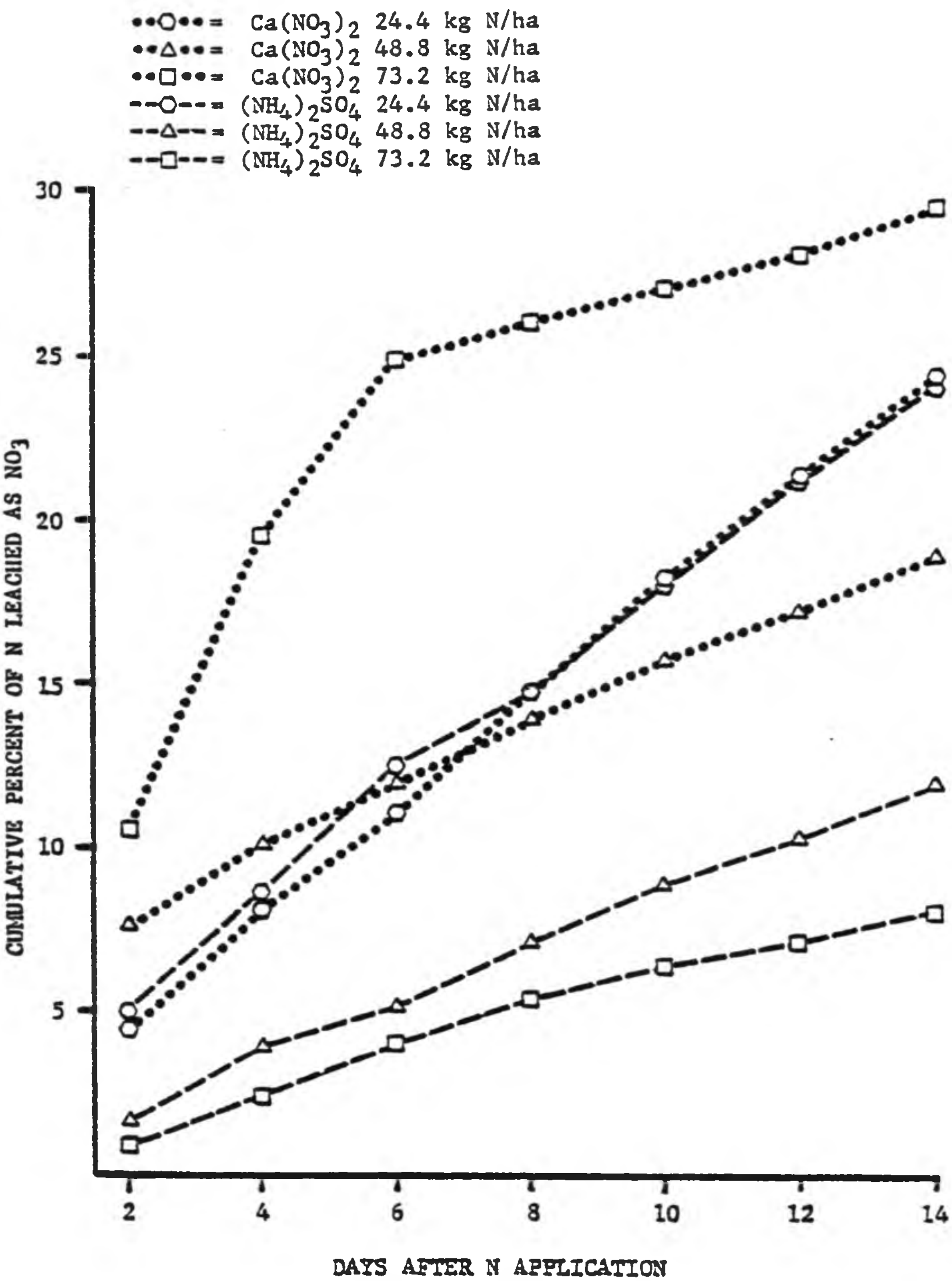

Figure 11. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the first 2-week cycle).

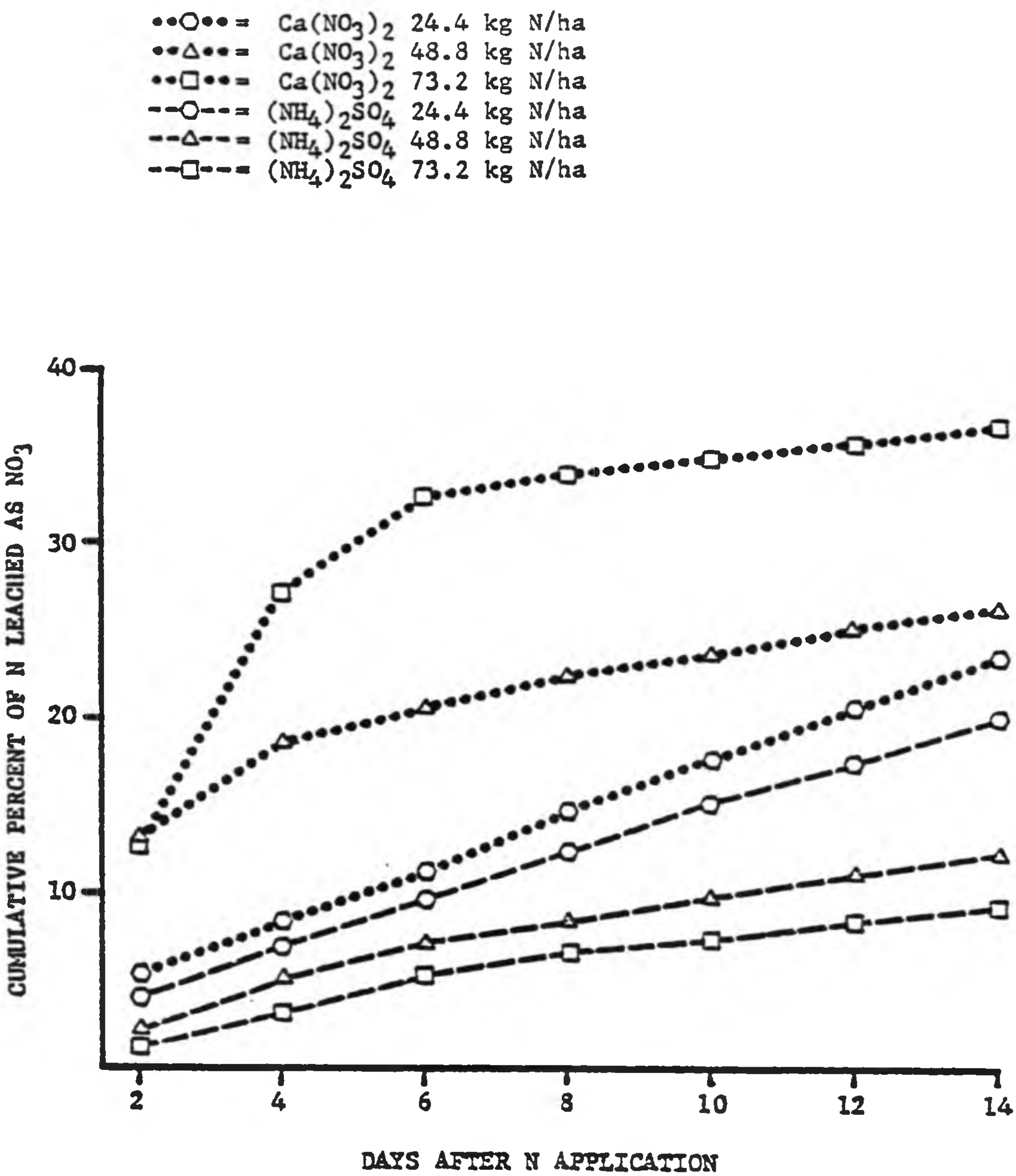

Figure 12. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the second 2-week cycle).

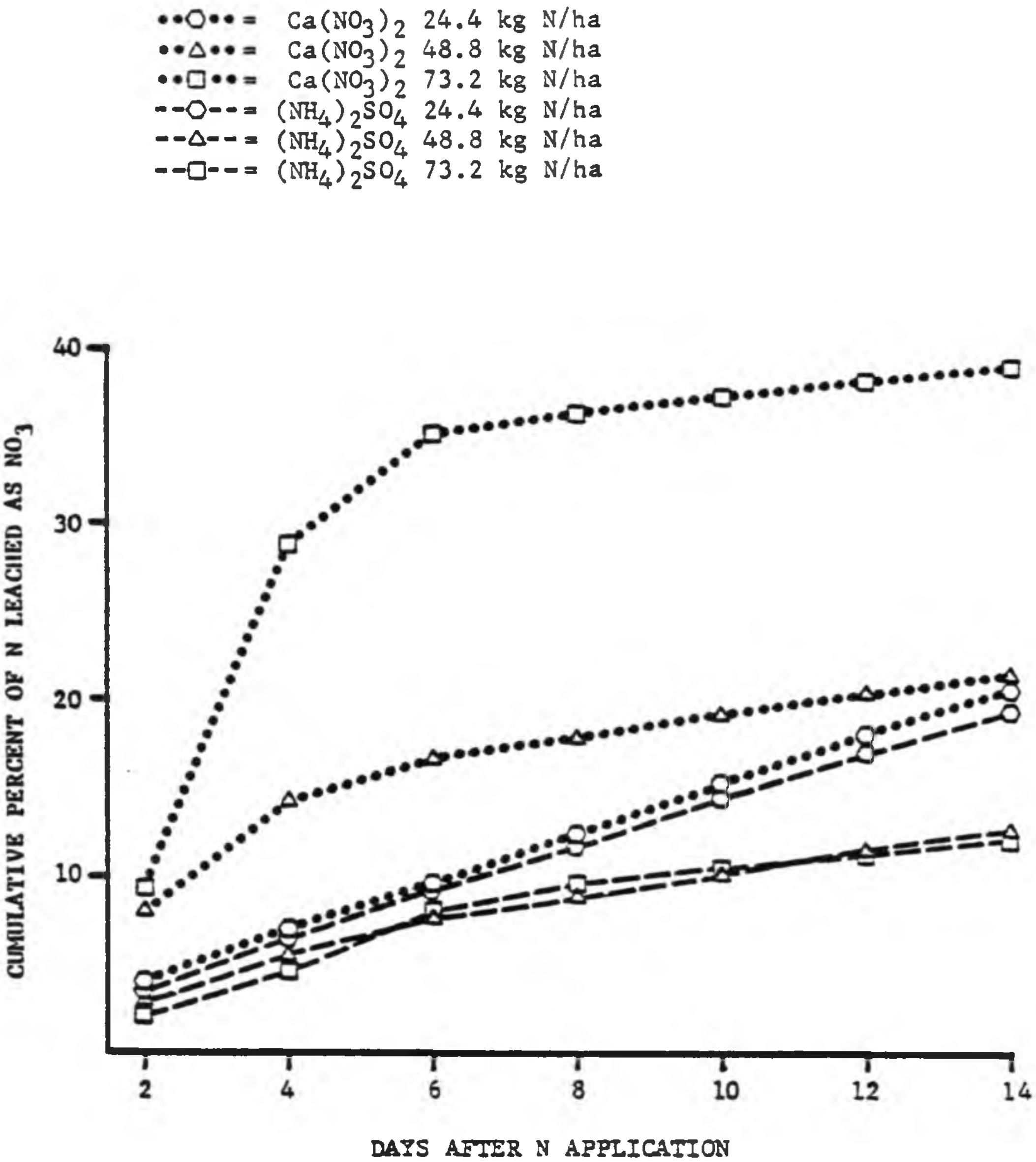

Figure 13. Percent of applied N leached as nitrate with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the third 2-week cycle).

determination methods. Since the nitrate determination was made by a "known addition" method (Orion Research Inc., 1973), it would seem likely that the total-N colorimetric method (Cataldo et al., 1974) was not as sensitive at low N levels as the nitrate method. Although statistical analysis was carried out on the data, results will be presented and discussed without reference to statistics.

In general, most of the total-N was leached during the first 6 days after N application with little difference after this (Table 8). The first 8 days show slightly greater total-N than NO_3-N readings indicating that most of the N leached was in the nitrate form. In all cases calcium nitrate leached more total-N than ammonium sulphate (Table 9). Nitrate moves much more freely with the soil solution than does ammonium however sand has little if any cation exchange capacity so ammonium sulphate would be expected to leach approximately the same quantity of total-N as calcium nitrate. One possible explanation could be that the ammonium-fed plants secreted hydroxyl ions in order to balance their internal pH which in turn, created a net negative charge on the surface of the roots. Since the root systems were very extensive, there may have been many sites where NH_4^+ ions could be held and therefore resist leaching. There were also greater amounts of total-N leached as N levels increased (Table 10), which is due mainly to calcium nitrate's influence in mean determination. Total-N leached

TABLE 8. EFFECT OF TIME IN DAYS ON TOTAL-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Total-N (kg/ha)		
Day	Cycle 1	Cycle 2	Cycle 3
2	3.13a*	3.78a	3.62a
4	1.75b	4.00a	4.33a
6	0.82c	1.70b	2.03b
8	0.33cd	0.77bc	0.55c
10	0.27cd	0.22c	0.27c
12	0.16d	0.11c	0.11c
14	0.16d	0.05c	0.05c

*For each column, means for treatment effects followed by the same letter do no differ significantly (BLSD=.05).

[w]Average of 2 N sources, 3 N levels, and 2 mycorrhizal levels.

TABLE 9. EFFECT OF N SOURCE ON TOTAL-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Total-N (kg/ha/leaching)		
N Source	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	1.43*	2.36*	2.19*
$(NH_4)_2SO_4$	0.44	0.66	0.93

*Means in the same column are significantly different ($p=0.05$).

[w]Average of 3 N levels, 2 mycorrhizal levels and 7 leaching dates.

TABLE 10. EFFECT OF N LEVEL ON TOTAL-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Total-N (kg/ha/leaching)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3
24.4	0.33c*	0.44c	0.33c
48.8	0.77b	1.21b	1.15b
73.2	1.70a	2.91a	3.18a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources, 2 mycorrhizal levels and 7 leaching dates.

TABLE 11. EFFECT OF N SOURCE AND N LEVEL ON TOTAL-N LEACHED AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Total-N (kg/ha/leaching)		
N Source	N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	0.38c*	0.55c	0.38c
	48.8	1.10b	1.81b	1.59b
	73.2	2.74a	4.71a	4.71a
$(NH_4)_2SO_4$	24.4	0.27c	0.33c	0.27c
	48.8	0.38c	0.60c	0.77c
	73.2	0.71bc	1.10bc	1.64b

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 mycorrhizal levels and 7 leaching dates.

at the end of each two week cycle was higher for calcium nitrate than for ammonium sulphate (Figures 14, 15, 16). In all cases, with increasing rates or calcium nitrate there were increasing amounts of total-N in the leachate. This trend held true for ammonium sulphate but to a lesser extent. Generally speaking, there were no differences between the ammonium sulphate rates except for the third cycle at the high rate. Differences between the calcium nitrate rates did occur (Table 11).

On a daily basis, total-N leached was highest at the high calcium nitrate rate with leveling off occurring after the sixth day (Figures 17, 18, 19). The medium rate of calcium nitrate leached the next greatest amount of total-N with leveling off occurring after the fourth day. In the third cycle, the high ammonium sulphate rate leached about the same quantity of total-N as the medium calcium nitrate rate.

With decreasing calcium nitrate rates, percentage N leached on a daily basis also decreased (Figures 20, 21, 22). In the third cycle, the total percentages N leached for the high rates of calcium nitrate and ammonium sulphate were 44.26% and 15.59% respectively. These are higher than the percentages NO_3-N leached suggesting that there may have

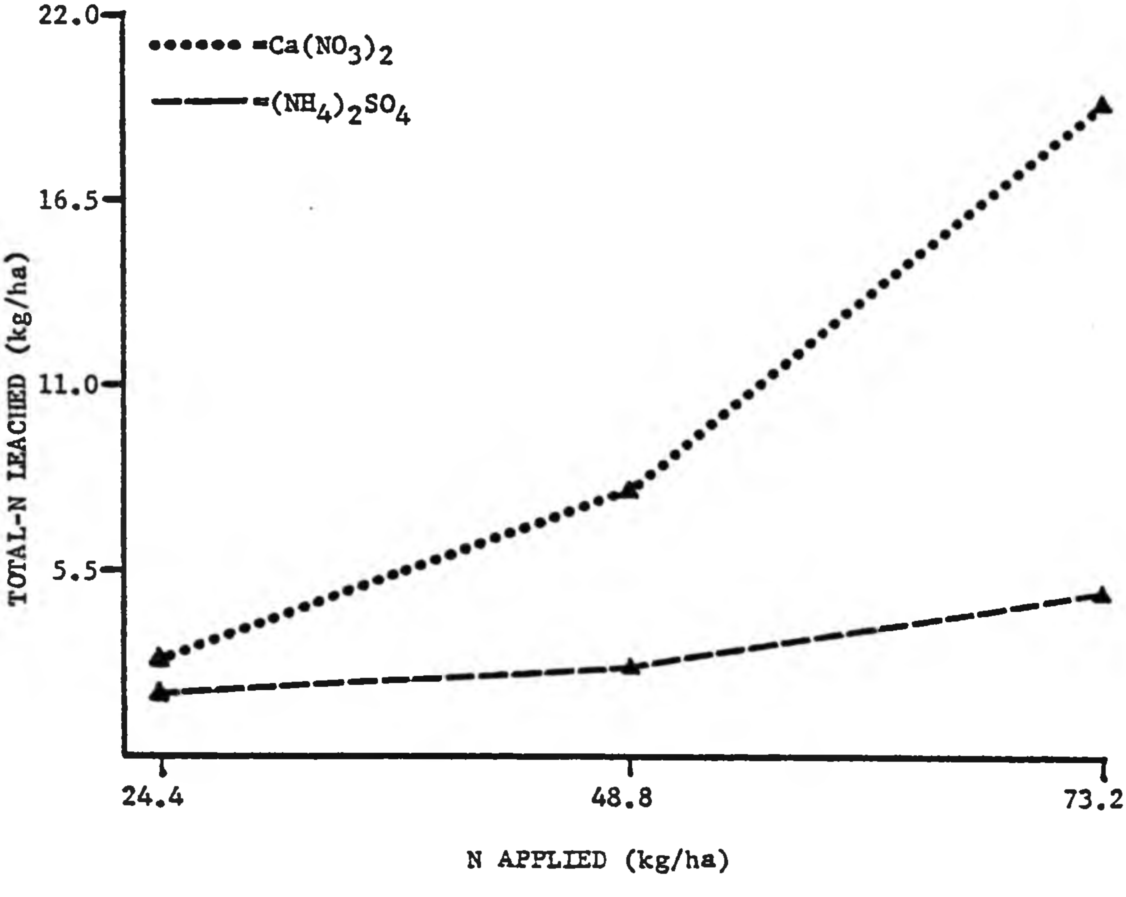

Figure 14. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the first 2-week cycle).

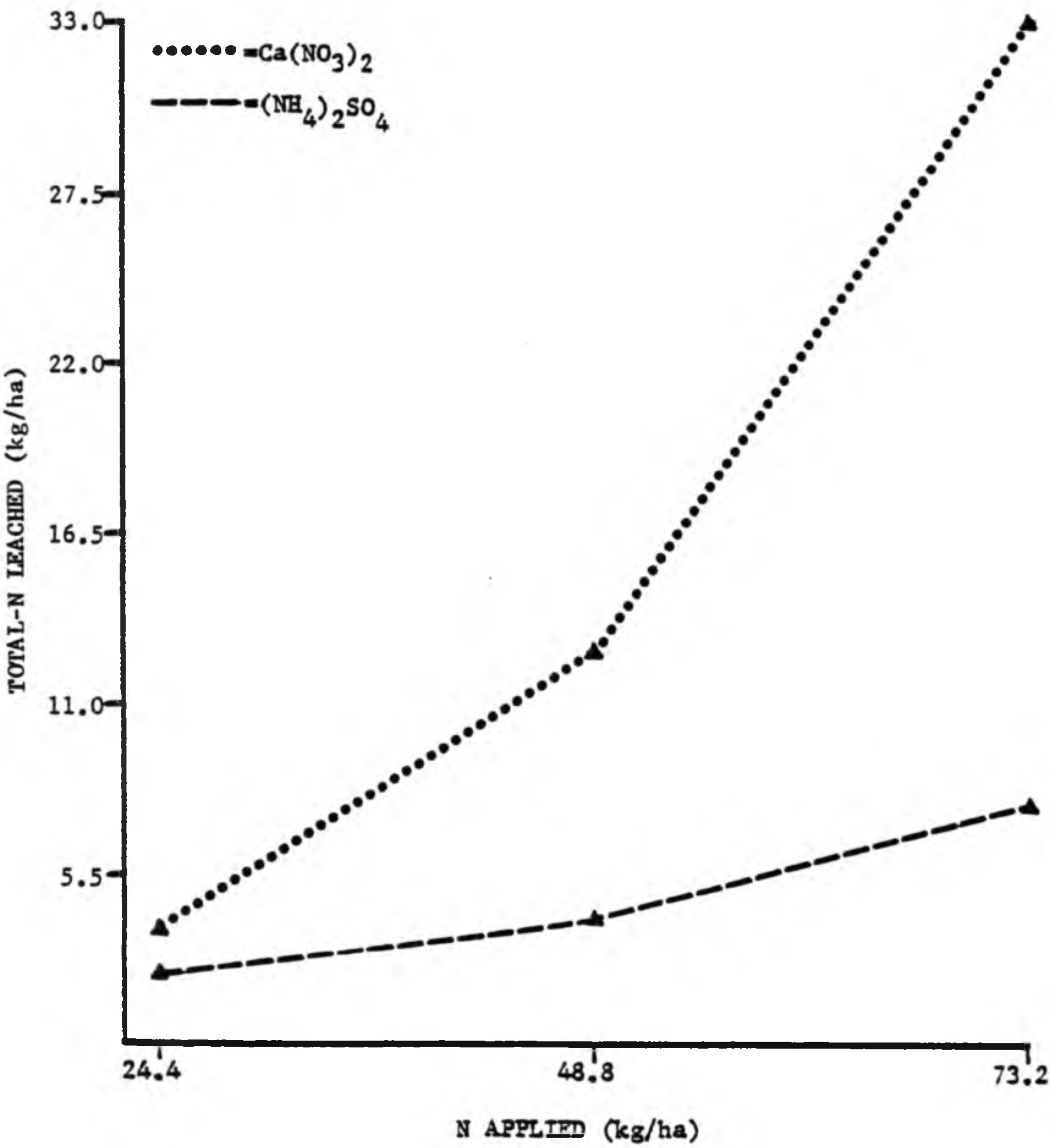

Figure 15. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the second 2-week cycle).

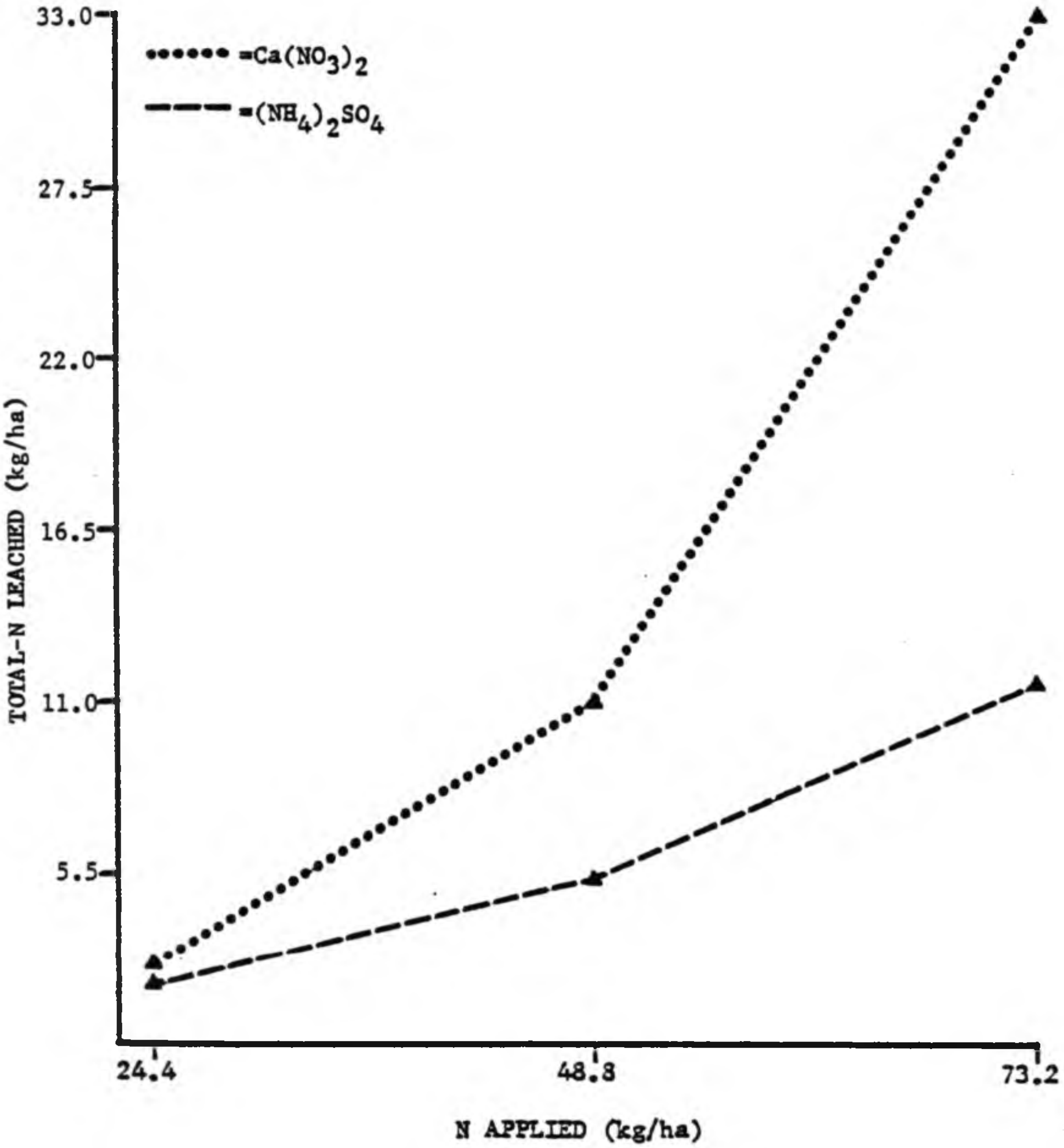

Figure 16. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (sum of 7 sampling dates for the third 2-week cycle).

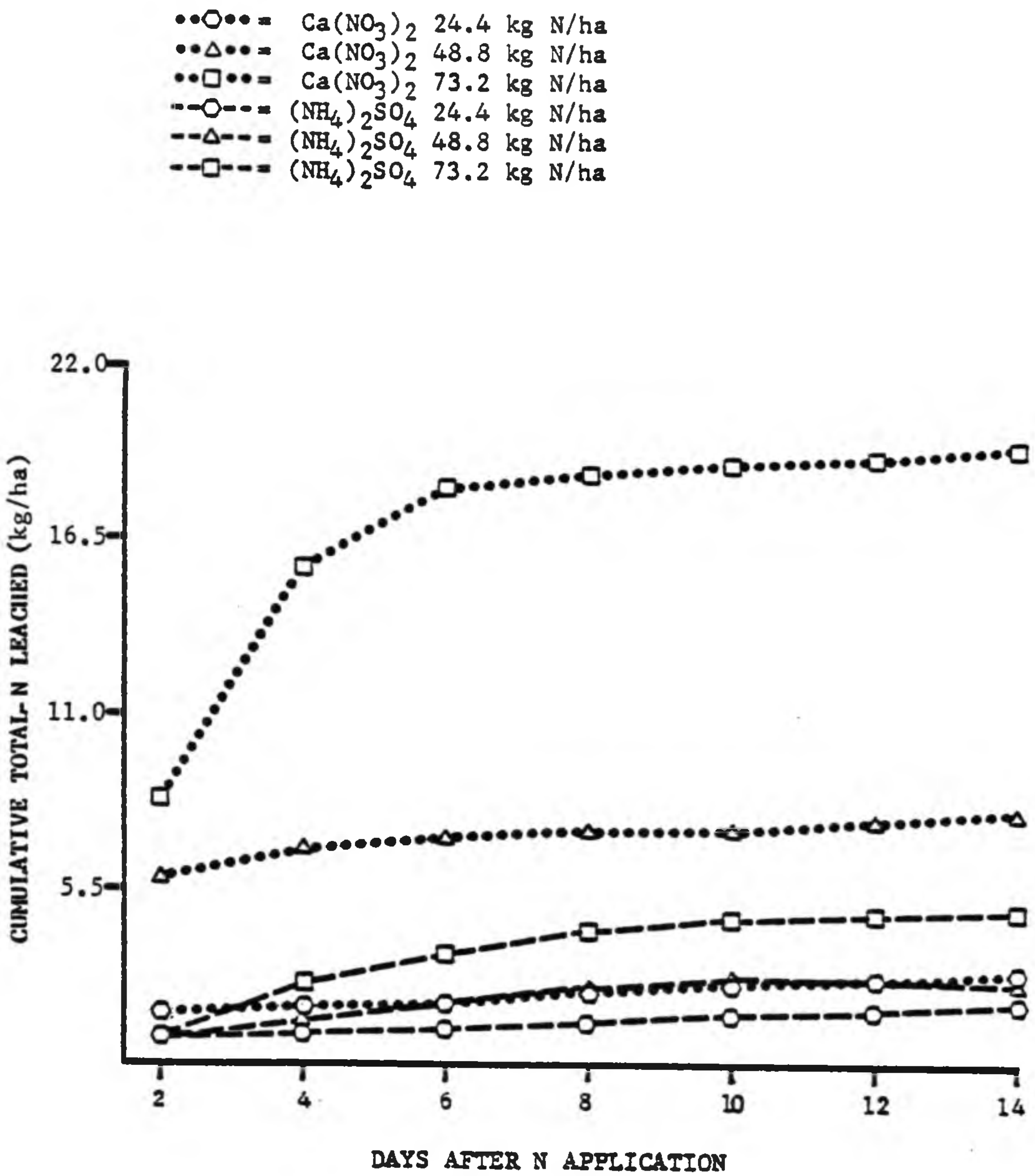

Figure 17. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the first 2-week cycle).

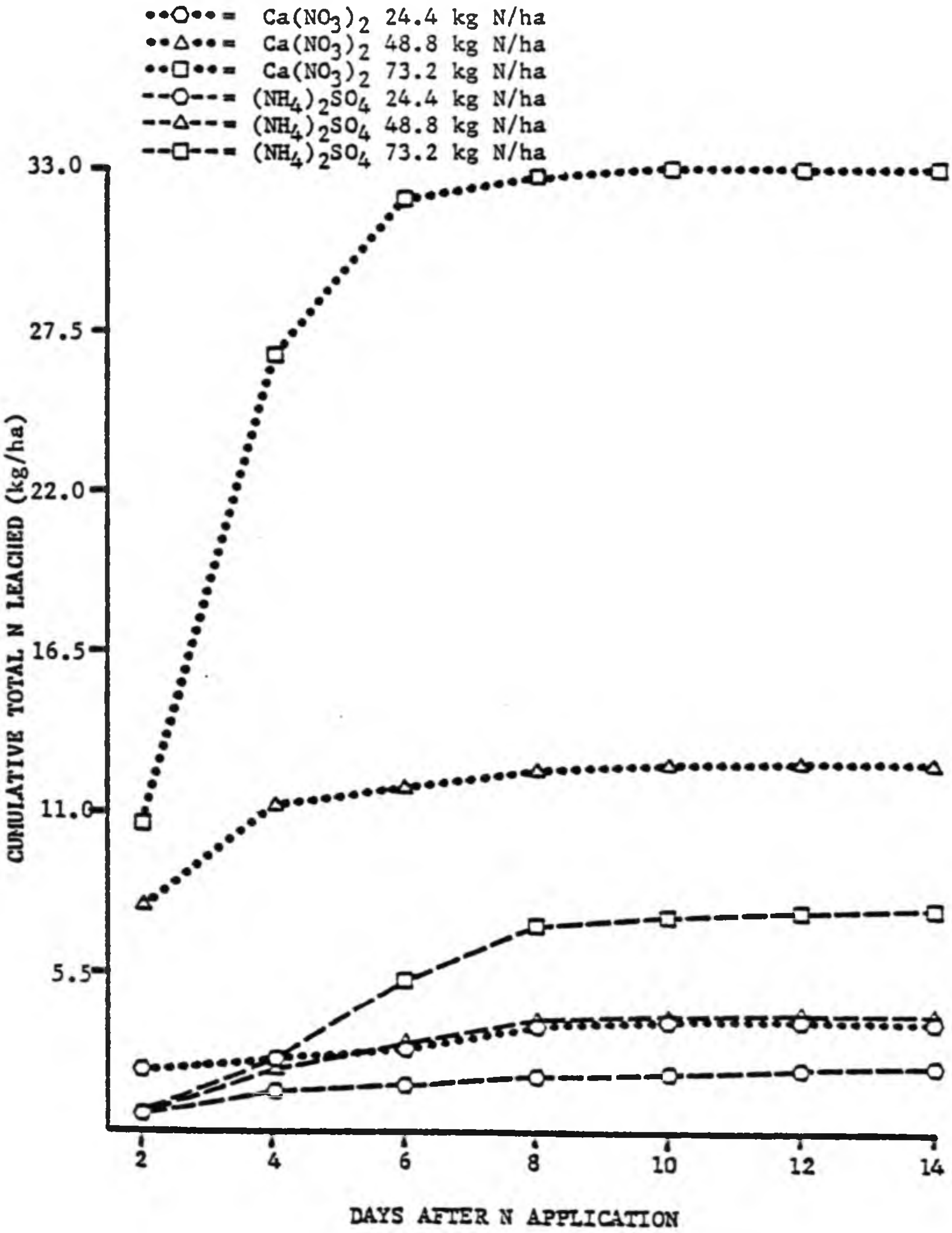

Figure 18. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the second 2-week cycle).

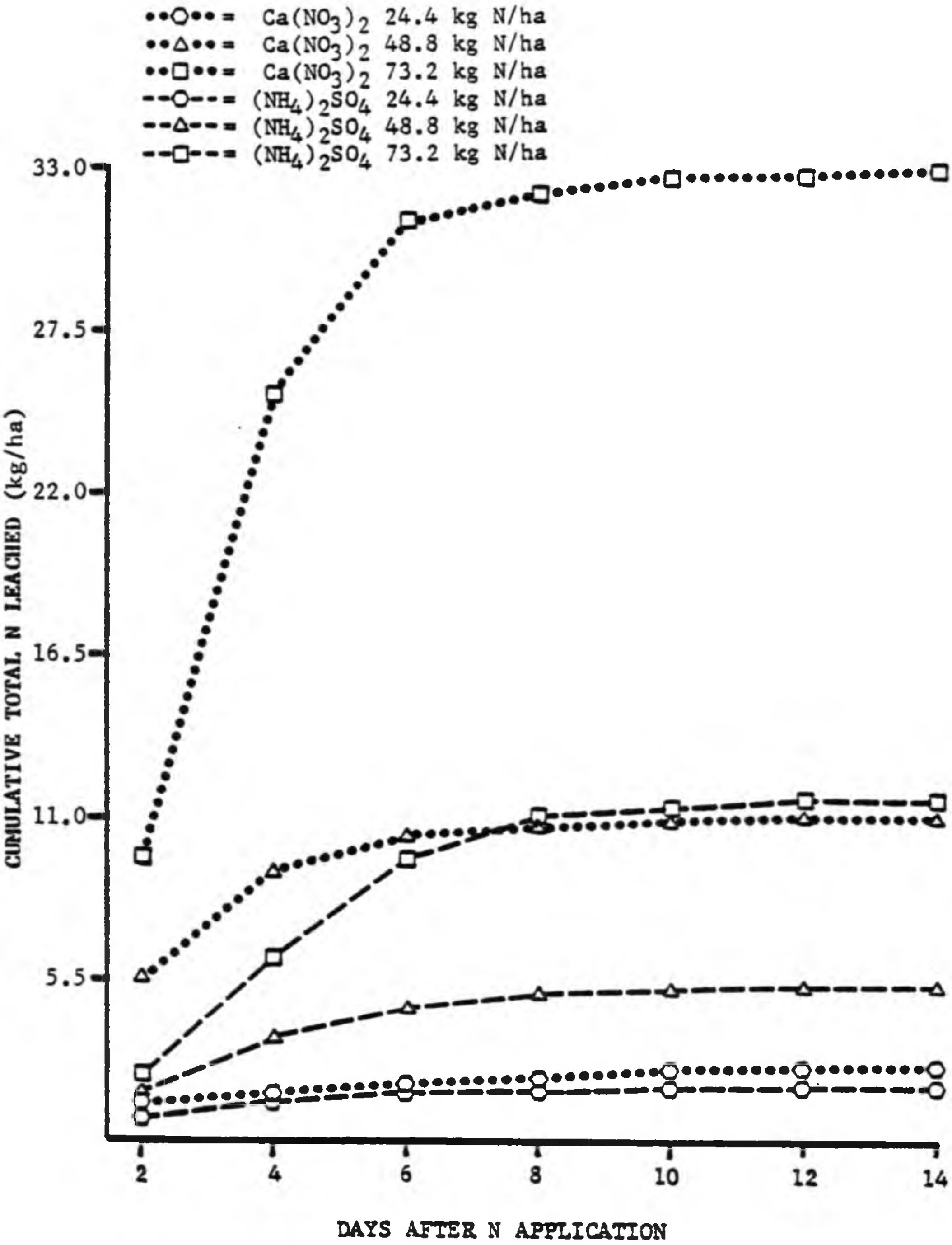

Figure 19. Total-N leached with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative kg N/ha leached at 7 sampling dates for the third 2-week cycle).

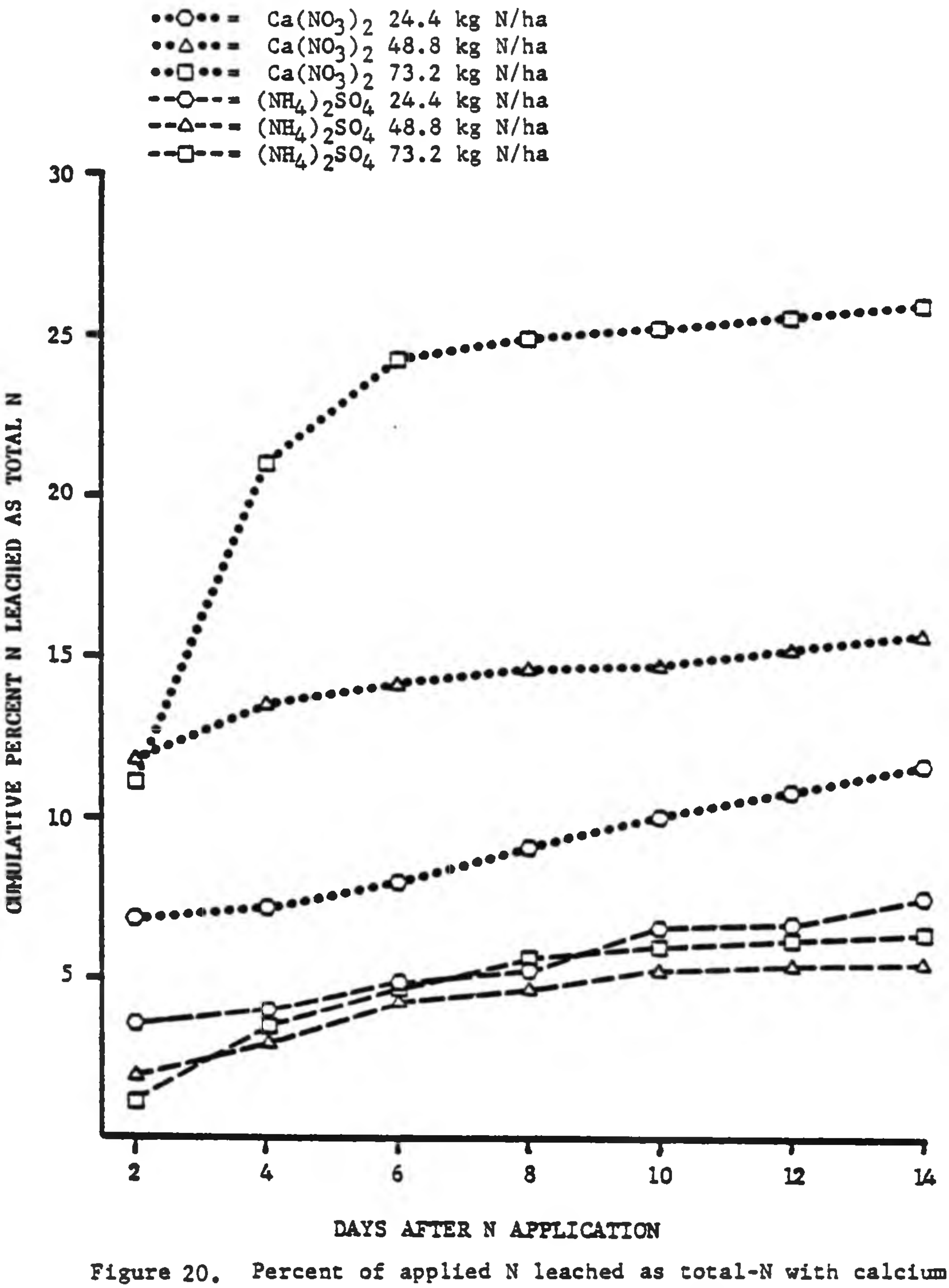

Figure 20. Percent of applied N leached as total-N with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the first 2-week cycle).

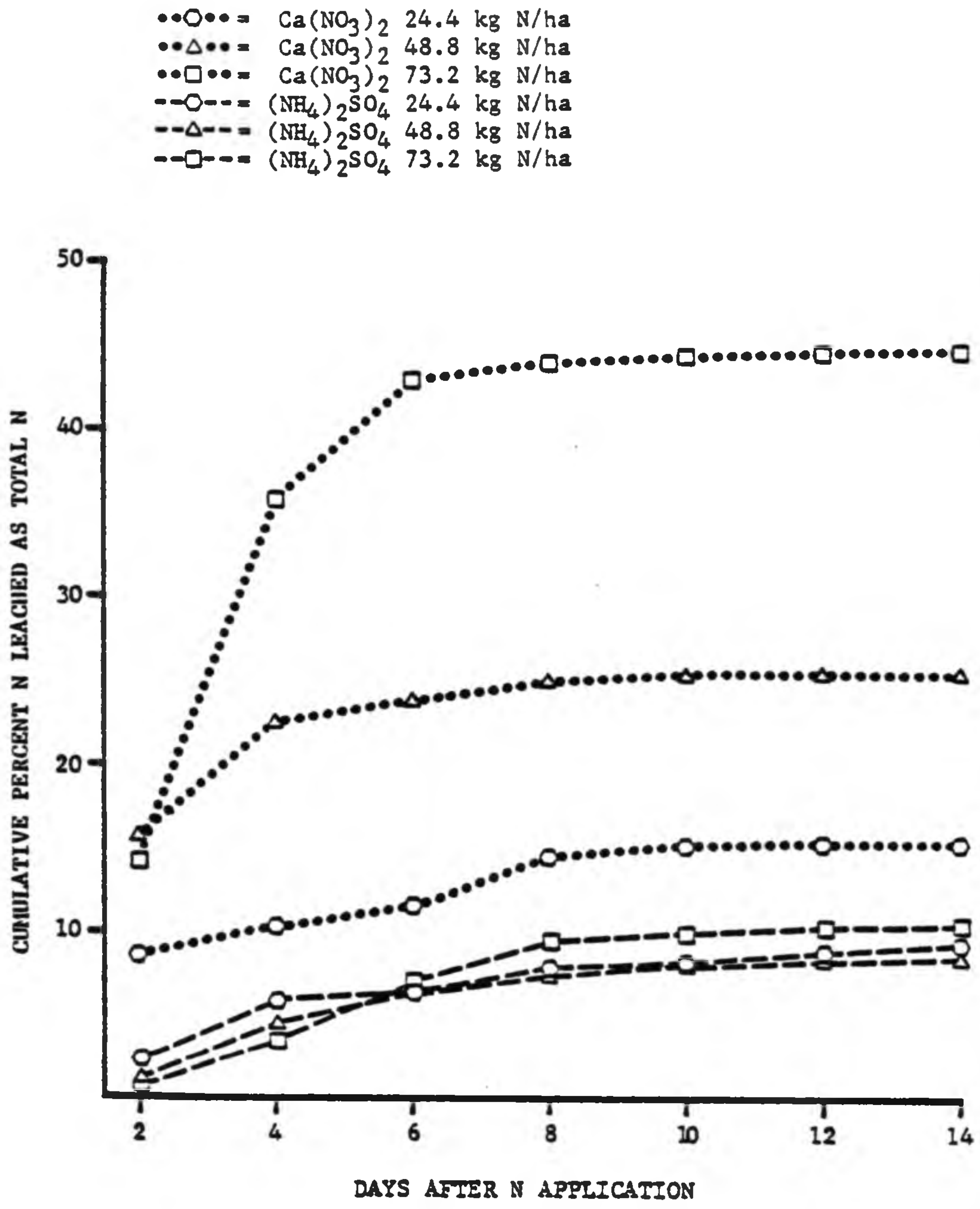

Figure 21. Percent of applied N leached as total-N with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the second 2-week cycle).

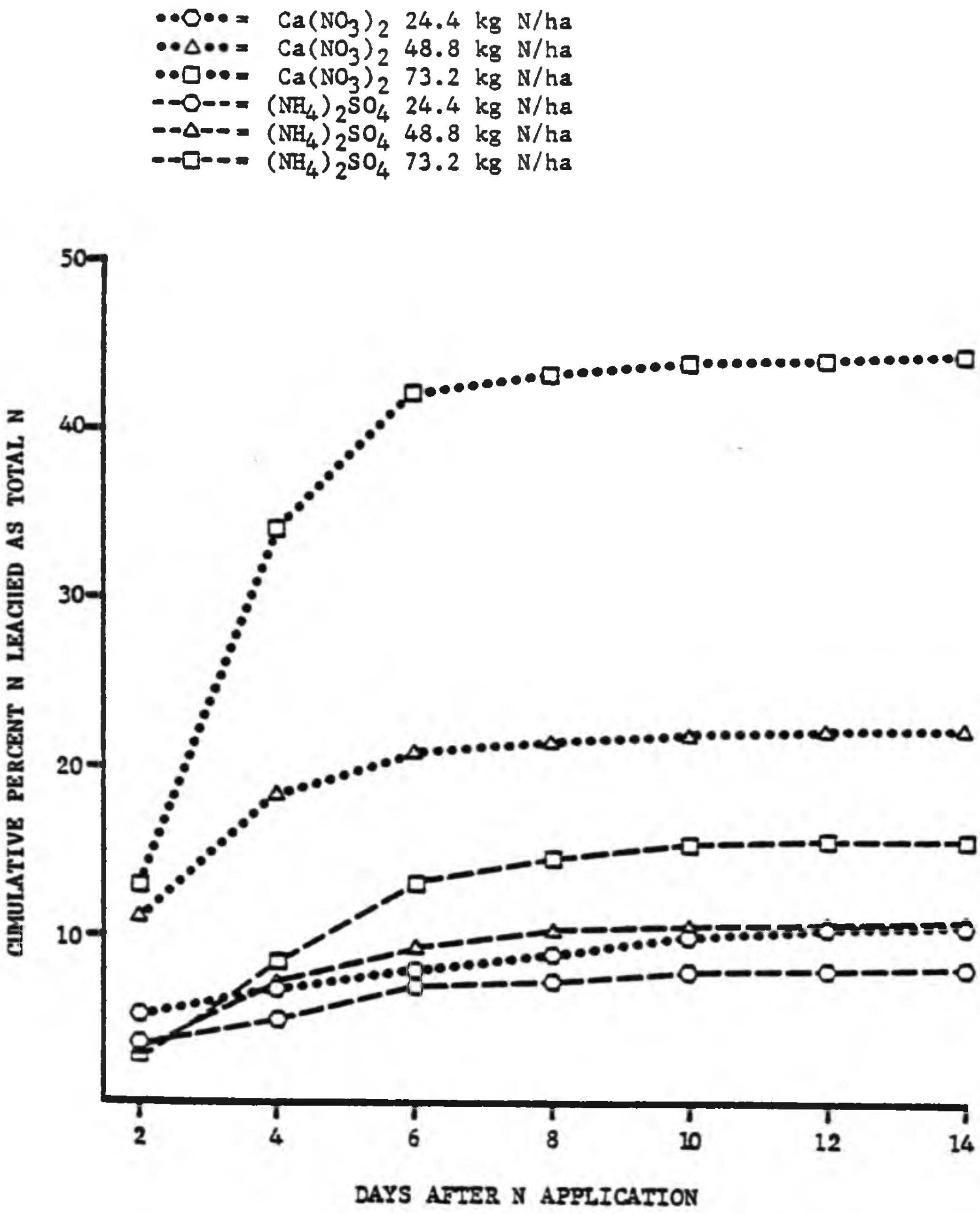

Figure 22. Percent of applied N leached as total-N with calcium nitrate and ammonium sulphate at 24.4, 48.8 and 73.2 kg N/ha (cumulative percent N leached at 7 sampling dates for the third 2-week cycle).

been other forms of N in the leachate such as ammonium, nitrite or organic N from decomposing roots and soil microorganisms.

There were no mycorrhizal effects on total-N leached.

III. VISUAL RATINGS

The response of common bermudagrass to treatments was apparent at the end of each two week cycle before clippings were removed.

Visual ratings were consistently higher for mycorrhizal plants, however, differences were not significant.

Mycorrhizal plants formed a denser and more uniform stand than nonmycorrhizal plants resulting in their higher ratings, however differences in color and general appearance were not noticeable thus the lack of a significant mycorrhizal effect. It should be noted that during the establishment phase, the plants were already showing a marked difference in density.

There was a significant (Anova, $p<.05$) N level effect in the third cycle with increasing visual ratings corresponding to increasing N levels (Table 12) and a significant interaction (Anova, $p<.05$) between N source and N level. Best visual ratings were found with the 73.2 kg N/ha ammonium sulphate rate, although it was not different from the 48.8 kg N/ha calcium nitrate rate ($p>0.05$) (Table 13). All other means were not significantly different.

TABLE 12. EFFECT OF N LEVEL ON VISUAL RATINGS[v] OF COMMON BERMUDAGRASS AT THE END OF THREE 2-WEEK CYCLES[w]

N Level (kg/ha)	Visual Rating		
	Cycle 1	Cycle 2	Cycle 3
24.4	2.82	2.89	2.70b*
48.8	2.98	3.01	2.91ab
73.2	2.89	3.03	3.13a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[v]Visual ratings on scale of 1 - 5 with 1 poorest and 5 best

[w]Average of 2 N sources, and 2 mycorrhizal levels

TABLE 13. EFFECT OF N SOURCE AND N LEVEL ON VISUAL RATINGS[v] OF COMMON BERMUDAGRASS AT THE END OF THREE 2-WEEK CYCLES[w]

N Source	N Level (kg/ha)	Visual Rating		
		Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	2.83	2.90	2.75b*
	48.8	3.06	3.13	3.05ab
	73.2	2.83	2.88	2.73b
$(NH_4)_2SO_4$	24.4	2.80	2.88	2.65b
	48.8	2.90	2.88	2.77b
	73.2	2.95	3.18	3.53a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[v]Visual ratings on scale of 1 - 5 with 1 poorest and 5 best

[w]Average of 2 mycorrhizal levels.

IV. CLIPPING DRY WEIGHTS

Clipping dry weights increased with increasing levels of ammonium sulphate, however, highest yields obtained with calcium nitrate were at the 48.8 kg N/ha rate (Table 14). Highest clipping dry weights corresponded to the highest visual ratings. Mean clipping dry weights were statistically the same for the 48.8 and 73.2 kg N/ha rate of ammonium sulphate and 48.8 kg N/ha calcium nitrate rate. There were no significant mycorrhizal effects. However, clipping dry weight means of inoculated plants were consistently higher than non-inoculated plants throughout the experiment.

V. CHLOROPHYLL CONTENTS

Calcium nitrate gave greater chlorophyll contents than ammonium sulphate throughout the experiment however there was no significance between means. Mycorrhizal means were lower than non-mycorrhizal with ammonium sulphate however the trend was reversed when calcium nitrate was the N source (Table 15).

VI. VERDURE DRY WEIGHTS

Verdure was harvested once at the end of the experiment. A significant positive effect (Anova, $p<.05$) of mycorrhiza on verdure dry weight occurred. Differences between means were significant (BLSD, $p<.05$) with mycorrhizal plants averaging 8838 kg/ha and nonmycorrhizal

TABLE 14. EFFECT OF N SOURCE AND N LEVEL ON CLIPPING, VERDURE AND ROOT DRY WEIGHTS[v]

		Clipping Dry Weight (kg/ha)				
N Source	N Level (kg/ha)	Cycle 1[w]	Cycle 2	Cycle 3	VDW[y] (kg/ha)	RDW[z] (kg/ha)
$Ca(NO_3)_2$	24.4	477b*	691b	598b	9057	15767ab
	48.8	685a	850ab	811ab	8284	11787b
	73.2	422b	751ab	647b	7922	12133b
$(NH_4)_2SO_4$	24.4	422b	663b	592b	8300	13745ab
	48.8	493b	729ab	724ab	7538	14786ab
	73.2	521ab	921a	943a	8476	17813a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[v]Average of 2 mycorrhizal levels

[w]Sampled at 3 consecutive 2-week cycles.

[y]VDW = Verdure Dry Weight

[z]RDW = Root Dry Weight

TABLE 15. EFFECT OF N SOURCE AND MYCORRHIZAE ON CHLOROPHYLL CONTENTS OF BERMUDAGRASS CLIPPINGS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		mg chlorophyll/g dry tissue		
N source	Mycorrhizae	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	+[y]	2.91	4.46	4.47
	-	2.84	3.95	4.24
$(NH_4)_2SO_4$	+	2.35	4.15	3.89
	-	2.91	4.19	4.52

[w]Average of 3 N levels.

[y]Mycorrhizae present.

averaging 7692 kg/ha. As was mentioned before, visual ratings were partially influenced by plant density. The greater verdure dry weights of mycorrhizal plants is an indication of increased plant density and this was in part due to effects during the establishment phase.

VII. ROOT DRY WEIGHTS

Root dry weights were measured once at the termination of the experiment. A highly significant (Anova, $p<.001$) positive mycorrhizal effect was noted. Differences between means were significant (BLSD, $p<.05$) with mycorrhizal plants averaging 18854 kg/ha and nonmycorrhizal plants averaging 9819 kg/ha. A significant interaction (Anova, $p<.05$) between N source and N rates was also apparent. With increasing levels of ammonium sulphate, there were corresponding increases in root dry weights while greatest root dry weights among the calcium nitrate means were found with the 24.4 kg N/ha rate (Table 11). N source effects showed a low level of significance (Anova, $p=.08$) with ammonium sulphate averaging 15450 kg/ha and calcium nitrate averaging 13229 kg/ha. Increases in root dry weight with mycorrhizal infection has been shown to occur in many studies (Schultz et al., 1981; Wallace, 1981; Wallace et al., 1982) with varying types of plants. The reason for this is not well known. One possibility could be improved plant nutrition. Another recent study (Barea and

Azcon-Aguilar, 1982) indicated that *G. mosseae* is capable of synthesizing at least two gibberellin-like substances, one with a R_f corresponding in position to gibberellic acid, and four substances with the properties of cytokinins.

The fact that root dry weights increased with increasing ammonium sulphate levels and that root dry weights were greater with ammonium sulphate than calcium nitrate indicates that there may have been preferential uptake of ammonium either by the host roots, hyphae of the VAM fungus, or both. The fact that VAM infection was also greater with ammonium sulphate than with calcium nitrate overall indicates a possible VAM fungus preference for ammonium..

VIII. TISSUE ANALYSIS (CLIPPINGS)

NITROGEN (N)

In all cases, percent and total uptake of N of turf clippings increased with increasing N rates (Table 16). There was also a significant mycorrhizal effect (Anova, $p<.01$) with mean percent N content of mycorrhizal plants significantly lower than nonmycorrhizal (Table 17). Mycorrhizal plants were also consistently lower than nonmycorrhizal in total uptake of N.

Increased N concentration in tissues of mycorrhizal plants has been consistently reported for ectotrophic and ericoid mycorrhizas (Hatch, 1937; Read and Stribley, 1973)

TABLE 16. EFFECT OF N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF N IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	percent N			Total N Uptake (kg/ha)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
24.4	1.99c[*]	2.02c	1.78c	12692b	13564b	10461b
48.8	2.24b	2.28b	2.06b	14627ab	17982a	15795a
73.2	2.47a	2.46a	2.21a	16151a	20499a	17637a

[*] For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w] Average of 2 N sources and 2 mycorrhizal levels.

TABLE 17. EFFECT OF MYCORRHIZAE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF N IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	percent N			Total N Uptake (kg/ha)		
Mycorrhizae	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
+[y]	2.19*	2.14*	1.95*	13268*	17242	14211
–	2.31	2.33	2.08	15707	17456	15049

*Means in the same column are significantly different (p=0.05).

[w]Average of 2 N sources and 3 N levels.

[y]Mycorrhizae present.

however results obtained with vesicular-arbuscular mycorrhizas are variable. Ross and Harper (1970) found higher N concentrations in leaf tissues of mycorrhizal soybean plants, suggesting an enhancement of N uptake brought about by mycorrhizae. Schenck and Hinson (1973) and Carling et al. (1978) argued against a direct effect of mycorrhiza in uptake of N since mycorrhizal infection did not usually increase N concentrations of nonlegumes, and that mycorrhiza did not increase growth of non-nodulating isolines of soybean under N-deficiency conditions. The importance of phosphorus in nodulation and N-fixation by Rhizobium spp. has been well documented (Von Schreven, 1958) and thus it appears that improved N nutrition of legumes is due to an enhancement of N-fixation due to increased hyphal translocation of phosphorus to the roots.

The lower N levels of mycorrhizal plants than non-mycorrhizal in the present investigation indicate possible competition between host and fungus. Certainly the fact that chlorophyll contents were generally lower for mycorrhizal plants when ammonium sulphate was the N source and no differences occurred in clipping dry weights between mycorrhizal and non-mycorrhizal plants support this hypothesis. Deficiencies of phosphorus may also be important. Salisbury (1975) conducted an experiment on tomato (Lycopersicum esculentum L.) plants and measured fresh and dry weights of tops in response to two different P levels (high and low) over a wide range of N concentrations. He found that as nitrogen increased, plants responded the same at both phosphorus levels until phosphorus became limiting at the nitrogen saturation level. He found that higher phosphorus led to a higher saturation level for nitrogen as evidenced by the greater fresh and dry weights of plants.

Growth depressions in mycorrhizal plants have been shown to occur when the soil phosphorus was not limiting to plant growth (Crush, 1973, 1976; Cooper, 1975). The physiology of growth depressions in mycorrhizal plants is not fully understood but may be related to carbon flow from the host to the fungus (Cox et al., 1975). Several workers have suggested that lipids in hyphae and spores of VAM fungi could cause a significant drain on the host carbohydrate supply (Ho and Trappe, 1973; Cox et al., 1975; Bevege et al., 1975; Cooper, 1975).

PHOSPHORUS (P)

There were no significant treatment effects on either P concentration or total uptake of P in clippings of common bermudagrass. Total uptake of P was greater with ammonium sulphate than with calcium nitrate, although differences between means were not significant at the 5% level (Table 18). Levels of P in turfgrass are said to be sufficient in

TABLE 18. EFFECT OF N SOURCE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF PHOSPHORUS IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent P			Total P Uptake (kg/ha)		
Nsource	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	.12	.11	.11	811	861	718
$(NH_4)_2SO_4$	.13	.12	.10	817	888	795

[w]Average of 3 N and 2 mycorrhizal levels

the 0.2 - 0.4% range (Fujimoto, 1980). Levels found in clippings in this experiment were extremely low ranging from 0.09 - 0.15%. These results indicate that the P level of the medium was extremely low since no extra P was added during the experiment and since it is well documented that improved P nutrition is the main beneficial effect of mycorrhizal infection. This factor probably plays a very important role in affecting all other parameters measured in this experiment.

P levels between mycorrhizal and non-mycorrhizal plants were not significantly different because turfgrass root systems are very fibrous and extensive and probably do not rely on mycorrhizal associations for P uptake to as great an extent as other plants with smaller root systems. This may partially account for lack of differences between mycorrhizal and non-mycorrhizal plants in P uptake in this experiment.

POTASSIUM (K)

Percent and total uptake of K was found to increase in clippings with increasing N levels (Table 19). Mean percent K readings were also found to be significantly greater in the second and third cycles when ammonium sulphate was used, however, differences between means were not significant, indicating a dilution effect (Table 20). The trend was still toward greater K readings when ammonium sulphate was

TABLE 19. EFFECT OF N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF K IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent K			Total K Uptake (kg/ha)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
24.4	1.66c*	1.55c	1.35b	10439b	10510b	7999b
48.8	1.79b	1.70b	1.49a	11733ab	13399a	11409a
73.2	1.96a	1.76a	1.50a	13015a	14830a	12209a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources and 2 mycorrhizal levels.

TABLE 20. EFFECT OF N SOURCE ON PERCENT AND TOTAL UPTAKE OF K IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent K			Total K Uptake (kg/ha)		
N Source	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	1.79	1.64*	1.40*	11491	12577	9677
$(NH_4)_2SO_4$	1.82	1.70	1.49	11968	13246	11404

*Means in the same column are significantly different (p=0.05).

[w]Average of 3 N and 2 mycorrhizal levels.

the N source. A significant interaction (Anova, $p<.01$) occurred between N source and N rate for percent K and total uptake of K in the second and third cycles (Table 21). Greatest percent K and total uptake was obtained with the 73.2 kg N/ha rate of ammonium sulphate and the 48.8 kg N/ha rate of calcium nitrate. Mean percent K was found to be significantly lower ($p<.05$) in mycorrhizal plants than non-mycorrhizal plants for the first two cycles (Table 22), however, significance was found only in the first cycle in the case of total K, suggesting that the lower percent K results were due to a dilution effect since mycorrhizal clipping dry weights were slightly higher than non-mycorrhizal. Although some studies have shown that mycorrhizae increase K concentrations in plants (Mosse, 1957; Baylis, 1959) the majority report significantly lower K concentrations with mycorrhizal as compared to non-mycorrhizal plants. Gerdemann (1964) Holevas (1966) Ross (1971) Deal et al. (1972) Kleinschmidt and Gerdemann (1972) all found increased total K uptake due to the larger size of the mycorrhizal plants. These variable results could reflect differences in K supply, the sodium:potassium ratio and N supply. These factors all influence internal concentration and distribution of cations in mycorrhizal plants (Chambers, Smith and Smith, 1980). The site of nitrate assimilation (root or shoot) in the plant species

TABLE 21. EFFECT OF N SOURCE AND N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF K IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Percent K			Total K Uptake (kg/ha)		
N source	N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	1.67	1.55c*	1.37bc	9847	10762c	8191cd
	48.8	1.78	1.70b	1.45b	12127	14496ab	11733b
	73.2	1.91	1.66b	1.38bc	12495	12473bc	9101bcd
$(NH_4)_2SO_4$	24.4	1.65	1.55c	1.32c	11031	10258c	7807d
	48.8	1.80	1.69b	1.54a	11338	12297bc	11086bc
	73.2	2.01	1.86a	1.62a	13536	17188a	15318a

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 mycorrhizal levels.

TABLE 22. EFFECT OF MYCORRHIZAE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF K IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent K			Total K Uptake (kg/ha)		
Mycorrhizae	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
+[y]	1.71*	1.63*	1.43	10521*	12862	10444
-	1.90	1.71	1.47	12939	12961	10636

*Means in the same column are significantly different (p=0.05).

[w]Average of 2 N sources and 3 N levels.

[y]Mycorrhizae present.

used, together with the mode of pH regulation, would also affect the distribution of cations within the plant (Smith, 1980).

With the results of this experiment, it seems that if top growth of mycorrhizal plants were significantly greater than non-mycorrhizal, there probably would have been greater total uptake of K. This did not occur however and is thought to be due to the extremely low P levels of the medium as indicated by the deficient levels found in both mycorrhizal and non-mycorrhizal treatments making for a less vigorous plant.

CALCIUM (Ca)

Percent Ca was found in general to increase in clippings with increasing rates of calcium nitrate and decrease with increasing rates of ammonium sulphate (Table 23). However with total uptake, trends for calcium nitrate and ammonium sulphate were generally the same since dry weights of clippings increased with increasing levels of ammonium sulphate.

Cox and Reisenauer (1973) using wheat as their indicator plant, showed that as nitrate uptake rates increased, so did Ca but with increasing rates of ammonium absorption, intake of calcium decreased. These higher rates of Ca intake with nitrate as opposed to ammonium may be attributed to reduced competition in the absorption process

TABLE 23. EFFECT OF N SOURCE AND N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Ca IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Percent Ca			Total Ca Uptake (kg/ha)		
N Source	N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	.49	.57ab*	.58b	2867	3876	3449
	48.8	.49	.56b	.57b	3290	4709	4605
	73.2	.54	.59ab	.62ab	3690	4419	4024
$(NH_4)_2SO_4$	24.4	.54	.64a	.66a	3618	4117	3832
	48.8	.51	.61ab	.62ab	3103	4441	4523
	73.2	.49	.54b	.55b	3092	4978	5208

*For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 mycorrhizal levels.

since Ca^{++} is a cation and nitrate is an anion while competition would exist between two cations. This seemed to be occurring to some extent, as seen with total uptake in the first cycle. However, with time, this trend did not occur which was possibly due to ammonium sulphate producing larger root systems thus giving a larger absorption surface.

MAGNESIUM (Mg)

Percent Mg and total uptake of Mg increased with increasing N levels (Table 24). This result is possibly due to a more vigorous plant at higher N levels thereby capable of greater uptake of Mg. Non-mycorrhizal percent Mg means were consistently higher than mycorrhizal means (Table 25) which could be due to a dilution effect since mycorrhizal clippings were slightly greater than non-mycorrhizal. There also seemed to be some competition occurring in the absorption process between ammonium and Mg since levels were generally lower with ammonium sulphate and since they are both cations (Table 26).

It is interesting to note that N, S and Mg content were all lower in mycorrhizal clippings than non-mycorrhizal since N and Mg are essential to the chlorophyll molecule and all are important in protein synthesis and composition again pointing to possible competition between host and fungus either for these elements directly or the products containing them.

TABLE 24. EFFECT OF N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Mg IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent Mg			Total Mg Uptake (kg/ha)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
24.4	.194b[*]	.214b	.216b	1195	1431b	1267b
48.8	.197b	.226ab	.228b	1288	1771a	1749a
73.2	.213a	.236a	.247a	1409	1968a	1968a

[*] For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w] Average of 2 N sources and 2 mycorrhizal levels.

TABLE 25. EFFECT OF MYCORRHIZAE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Mg IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent Mg			Total Mg Uptake (kg/ha)		
Mycorrhizae	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
+[y]	.19[*]	.21[*]	.22	1184[*]	1678	1678
-	.21	.24	.24	1409	1771	1639

[*] Means in the same column are significantly different (p=0.05).

[w] Average of 2 N sources and 3 N levels.

[y] Mycorrhizae present.

TABLE 26. EFFECT OF N SOURCE ON PERCENT AND TOTAL UPTAKE OF Mg IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent Mg			Total Mg Uptake (kg/ha)		
N Source	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	.20	.23	.24	1316	1749	1617
$(NH_4)_2SO_4$	.20	.22	.22	1277	1694	1705

[w]Average of 3 N and 2 mycorrhizal levels.

SULFUR (S)

Percent S and total uptake means were consistently higher for ammonium sulphate than calcium nitrate (Table 27). This stands to reason since S is being added with the ammonium sulphate treatments where ammonium sulphate is about 24% S and since it does not compete with ammonium in the absorption process. Total uptake of S increased with increasing levels of N (Table 28) which is probably due to a more vigorous plant. Percent S means were found to be lower for mycorrhizal than non-mycorrhizal treatments, however, with total uptake, S was not found to be significantly different in the second and third cycles (Table 29). This suggests that there was a dilution effect involved, since mycorrhizal clipping dry weights were slightly greater than non-mycorrhizal. Enhanced S uptake has been shown to occur with mycorrhiza (Gray and Gerdemann, 1973; Bowen et al., 1974). However, no conclusive evidence can be drawn from this experiment.

Silicon (Si)

In general, with increasing N levels, percent Si in clippings decreased (Table 30). This trend held true for both ammonium sulphate and calcium nitrate (Table 31) and also in the presence and absence of mycorrhiza (Table 32). When mycorrhizal means are compared to non-mycorrhizal (Table 33) percent Si is significantly greater than

TABLE 27. EFFECT OF N SOURCE ON PERCENT AND TOTAL UPTAKE OF S IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

N Source	Percent S			Total S Uptake (kg/ha)		
	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	.29*	.31*	.29*	1859	2347	1979
$(NH_4)_2SO_4$	.30	.33	.32	1952	2538	2385

*Means in the same column are significantly different (p=0.05).

[w]Average of 3 N and 2 mycorrhizal levels.

TABLE 28. EFFECT OF N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF S IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent S			Total S Uptake (kg/ha)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
24.4	.29	.32	.30	1820	2144b[*]	1787b
48.8	.30	.32	.30	1908	2473ab	2281ab
73.2	.30	.32	.31	1985	2703a	2489a

[*]For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources and 2 mycorrhizal levels.

TABLE 29. EFFECT OF MYCORRHIZAE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF S IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent S			Total S Uptake (kg/ha)		
Mycorrhizae	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
+[y]	.28*	.31*	.30	1749*	2440	2188
-	.31	.33	.31	2061	2440	2177

*Means in the same column are significantly different (p=0.05).

[w]Average of 2 N sources and 3 N levels.

[y]Mycorrhizae present.

TABLE 30. EFFECT OF N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Si IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

	Percent Si			Total Si Uptake (kg/ha)		
N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
24.4	1.17a[*]	1.22a	1.23a	7341	8196	7253
48.8	1.13ab	1.15b	1.17ab	7281	9057	9013
73.2	1.09b	1.07c	1.08b	7133	8887	8553

[*]For each column, means for treatment effects followed by the same letter do not differ significantly (BLSD=.05).

[w]Average of 2 N sources and 2 mycorrhizal levels.

TABLE 31. EFFECT OF N SOURCE AND N LEVEL ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Si IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Percent Si			Total Si Uptake (kg/ha)		
N Source	N level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
$Ca(NO_3)_2$	24.4	1.16	1.23	1.21	6809	8416	7231
	48.8	1.11	1.15	1.20	7604	9770	9753
	73.2	1.12	1.11	1.12	7281	8295	7248
$(NH_4)_2SO_4$	24.4	1.17	1.21	1.24	7878	7983	7281
	48.8	1.15	1.14	1.13	6957	8339	8273
	73.2	1.06	1.03	1.05	6985	9485	9868

[w]Average of 2 mycorrhizal levels.

TABLE 32. EFFECT OF MYCORRHIZAE AND N LEVEL ON PERCENT AND TOTAL UPTAKE OF Si IN CLIPPINGS OF COMMON BERMUDAGRASS AT 3 CONSECUTIVE 2-WEEK CYCLES[w]

		Percent Si			Total Si Uptake (kg/ha)		
Mycorrhizae	N Level (kg/ha)	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3
+[y]	24.4	1.19	1.24	1.26	8004	9194	8004
	48.8	1.18	1.23	1.24	7018	9616	9726
	73.2	1.14	1.12	1.11	6760	9243	8224
-	24.4	1.14	1.20	1.19	6678	7204	6502
	48.8	1.08	1.07	1.10	7544	8492	8306
	73.2	1.05	1.02	1.06	7511	8536	8887

[w]Average of 2 N sources.

[y]Mycorrhizae present.

TABLE 33. EFFECT OF MYCORRHIZAE ON PERCENT COMPOSITION AND TOTAL UPTAKE OF Si IN CLIPPINGS AT 3 CONSECUTIVE 2-WEEK CYCLES AND ROOTS OF COMMON BERMUDAGRASS[w]

	Clippings						Roots	
	Percent Si			Total Si Uptake (kg/ha)				Total
Mycorrhizae	Cycle 1	Cycle 2	Cycle 3	Cycle 1	Cycle 2	Cycle 3	% Si	Si Uptake (kg/ha)
+[y]	1.17*	1.20*	1.20*	7259	9353*	8651	1.36*	249260*
-	1.09	1.09	1.12	7242	8076	7900	.65	61491

*Means in the same column are significantly different ($p=0.05$).

[w]Average of 2 N sources and 3 N levels.

[y]Mycorrhizae present.

non-mycorrhizal although with total uptake, only the second cycle was significant. The fact that clipping dry weights and P levels of mycorrhizal and non-mycorrhizal plants were statistically the same in the presence or absence of mycorrhiza and since P was very deficient in both indicates that P levels in the medium were so low that mycorrhizal infection did not increase top growth so differences in total Si uptake did not show up too well.

Another factor influencing Si levels in the clippings is that with increasing levels of N, there was a decrease in Si content. This is thought to be due to a decrease in VAM infection with high levels of N which was found to occur especially with ammonium sulphate. Root tissue analysis data strongly suggests increased uptake of Si by mycorrhizal plants.

It has not been firmly established that mycorrhizae aid in Si uptake, however, a recent article by Yost and Fox (1982) and the results of this experiment indicate that this may be so. Yost and Fox found that Si contents of soybean plants from non-fumigated plot were usually twice that of plants from fumigated plots. They also found that with increasing levels of phosphorus above 0.2 mg P/liter, differences in Si content between mycorrhizal and non-mycorrhizal soybean plants became smaller indicating lower infection levels of mycorrhizal fungi or antagonistic effects of P on Si. They concluded that since mycorrhiza does substantially affect P uptake and the chemical properties of phosphate and silicate are so similar, mycorrhizal uptake of Si was probable.

Other studies indicate that silica gel is the most prevalent form of silicon in plants and that once it is solidified and deposited, it becomes immobile and cannot be translocated to other parts of the plant in the event of a Si deficiency at a later time (Yoshida et al., 1962). Thus mycorrhizae would be of importance in this case.

Okuda and Takahashi (1965) showed that with rice, both roots and shoots were longer in the presence of silicon and the grain yield was greater. They also found that iron and manganese toxicity was alleviated in the presence of silicon and that this was due to a decrease in the uptake of iron and manganese by the plants. They demonstrated that Fe^{++} and Mn^{++} were readily oxidized by rice roots, rendered insoluble, and precipitated on the surface of the roots.

In the present study, Si was not limiting in any treatments as the medium contains 49% (by weight) SiO_2 (Macdonald and Abbott, 1970). However, Mn contents of mycorrhizal roots were much greater than non-mycorrhizal roots. This trend was not found with Fe.

The possibility of contamination of the roots by particles of the medium after harvesting is ever present. Mycorrhizal hyphae have been shown to aid in aggregation of soil particles (Sutton and Shepard, 1976; Tisdall and Oades, 1979). It was shown that the hyphae grown in sand were covered with a layer of amorphous material, possibly polysaccharide, to which particles adhered. If contamination of the roots with soil particles were responsible for the increased Si content of mycorrhizal roots, it would be safe to assume that elemental analysis of mycorrhizal roots would show a greater concentration of Al and Fe, which did not occur (Table 34). With this evidence and that of Yost and Fox, it seems probable that mycorrhizae do enhance Si uptake.

TABLE 34. EFFECT OF MYCORRHIZAE ON CONCENTRATION OF Al AND Fe IN ROOTS OF COMMON BERMUDAGRASS AT THE FINAL HARVEST[w]

	ppm	
Mycorrhizae	Al	Fe
+[y]	1989	729*
-	2160	1175

*Means in the same column are significantly different ($p=0.05$).

[w]Average of 2 N sources and 3 N levels.

[y]Mycorrhizae present.

CHAPTER V

SUMMARY AND CONCLUSIONS

At the inception of this experiment, the effect of mycorrhizae on retention of N in golf greens constructed of sand was considered, since one study (Haines and Best, 1976) showed that mycorrhizae significantly reduced both NH_4-N and NO_3-N losses from forest soils. The implications of these results were great, since N management of high quality turfgrass is so intense and also because golf greens, after being constructed are nearly always fumigated with methyl bromide which destroys mycorrhizal populations as well as all other soil microorganisms. The other important factor is that golf greens are constructed of very sandy soil mixtures over a gravel and tile drainage system and are subjected to heavy irrigation schedules creating a high potential for loss of applied N through leaching.

The results of the present experiment, however, did not indicate mycorrhizal influence on downward movement of N. Possible factors involved were 1) the levels of N applied in quality turfgrass and in this experiment are quite high so that if the ambient N concentration of the medium exceeded the concentration at which maximum N uptake occurred, the reduction of soil solution N concentrations would not be evident as it might have been at lower ambient concentrations 2) the soluble forms of N used, despite their advantages, immediately become part of the soil solution which increases the chance of N-loss to downward movement of water and 3) the highly porous medium and heavy leaching schedule caused rapid N movement through and out of the root zone. Thus, under conditions of this experiment and of golf greens in general, there does not seem to be an effect of mycorrhizae on downward movement of N.

VAM infection levels decreased with increasing levels of ammonium sulphate but remained the same for calcium nitrate which is thought to be partly due to the high leaching characteristics of nitrate. VAM levels also decreased with increasing depths. Higher VAM infection levels with ammonium sulphate indicate a possible preference of G. mosseae for ammonium over nitrate.

In comparing the two sources of nitrogen used in this experiment and their relative movement through crushed basalt, calcium nitrate moved more rapidly and lost more N to leaching than did ammonium sulphate. N lost as nitrate averaged over three 2-week periods was 23%, 23%, and 35% of N applied for the 24.4, 48.8 and 73.2 kg N/ha rates respectively of cacium nitrate. This represents a considerable loss to the turf manager especially with rising costs of fertilizer. Ammonium sulphate losses were 21%, 12% and 10% for the 24.4, 48.8 and 73.2 kg N/ha rates, respectively indicating much more efficient use especially at the 48.8 and 73.2 kg N/ha rates of application.

These results occurred under conditions of excessive leaching and could be controlled by regulating the amount of water applied so as to minimize leachate volumes. Generally, studies show that the least amount of N was lost from the root zone when heavy irrigations were applied infrequently. In general, the 24.4 kg N/ha rate of calcium nitrate leached about the same amount of NO_3-N as did the three rates of ammonium sulphate.

With respect to visual ratings, best results were obtained with ammonium sulphate at the 73.2 kg N/ha rate although the 48.8 kg N/ha rate of calcium nitrate was statistically comparable. These differences, however, did not manifest themselves until the sixth week. Highest ratings correspond to greatest clipping dry weights, chlorophyll contents and tissue N composition. Mycorrhizae did not greatly influence visual ratings.

Chlorophyll contents were generally lower in mycorrhizal plants than non-mycorrhizal when ammonium sulphate was the N source but the trend was reversed with calcium nitrate. This along with significantly greater VAM infection levels with ammonium sulphate and lower N levels of clippings in mycorrhizal plants indicate possible competition between host and fungus for NH_4-N.

Clipping dry weights were greatest with the 73.2 kg N/ha rate of ammonium sulphate followed by the 48.8 kg N/ha calcium nitrate rate. Mycorrhizal infection did not increase clipping dry weights which indicates possible growth depressions due to competition between fungus and host for carbon sources. Low P levels may have also influenced clipping dry weights.

Verdure dry weights were not affected by treatments since verdure was a product of the pre-experiment, establishment phase.

Root dry weights decreased with increasing rates of calcium nitrate but the reverse was true with ammonium sulphate. Normally, there is a decrease in root weights with increasing N levels due to priority of the photosynthetic area for assimilates. However with ammonium sulphate, increases in root dry weight with increasing N levels may be due to the fact that ammonium can be metabolized in the roots thus making the organic compounds produced readily available to the root cells for growth whereas nitrate is usually translocated to the shoots where it is metabolized in the chloroplasts. Root dry weights were also greater in mycorrhizal as compared to non-mycorrhizal plants.

With increasing levels of N, there were correspondingly increasing concentrations of N, K and Mg in clippings. Ca was also found to increase but only when calcium nitrate was the source of N. Ca concentrations decreased with increasing ammonium sulphate levels. Si also exhibited a decreasing trend with both sources of N indicating a possible relationship with succulence of the plant. Possibly, as the plant becomes less succulent in response to lower N levels, it increases Si content for greater structural rigidity and hardiness. There also seemed to be enhanced uptake of Si by mycorrhizal fungi as evidenced by higher concentrations in roots and shoots of infected plants. P and S concentrations remained unchanged with N levels.

Ca concentrations were higher in plants receiving calcium nitrate while S and K were higher with ammonium sulphate. No differences were noted for the other elements. Because of the interacting factors of ionic competition, soil properties, mycorrhizal influence and low soil P levels, it is difficult to make any assumptions as to the reasons for differences obtained in elemental composition of turfgrass clippings.

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